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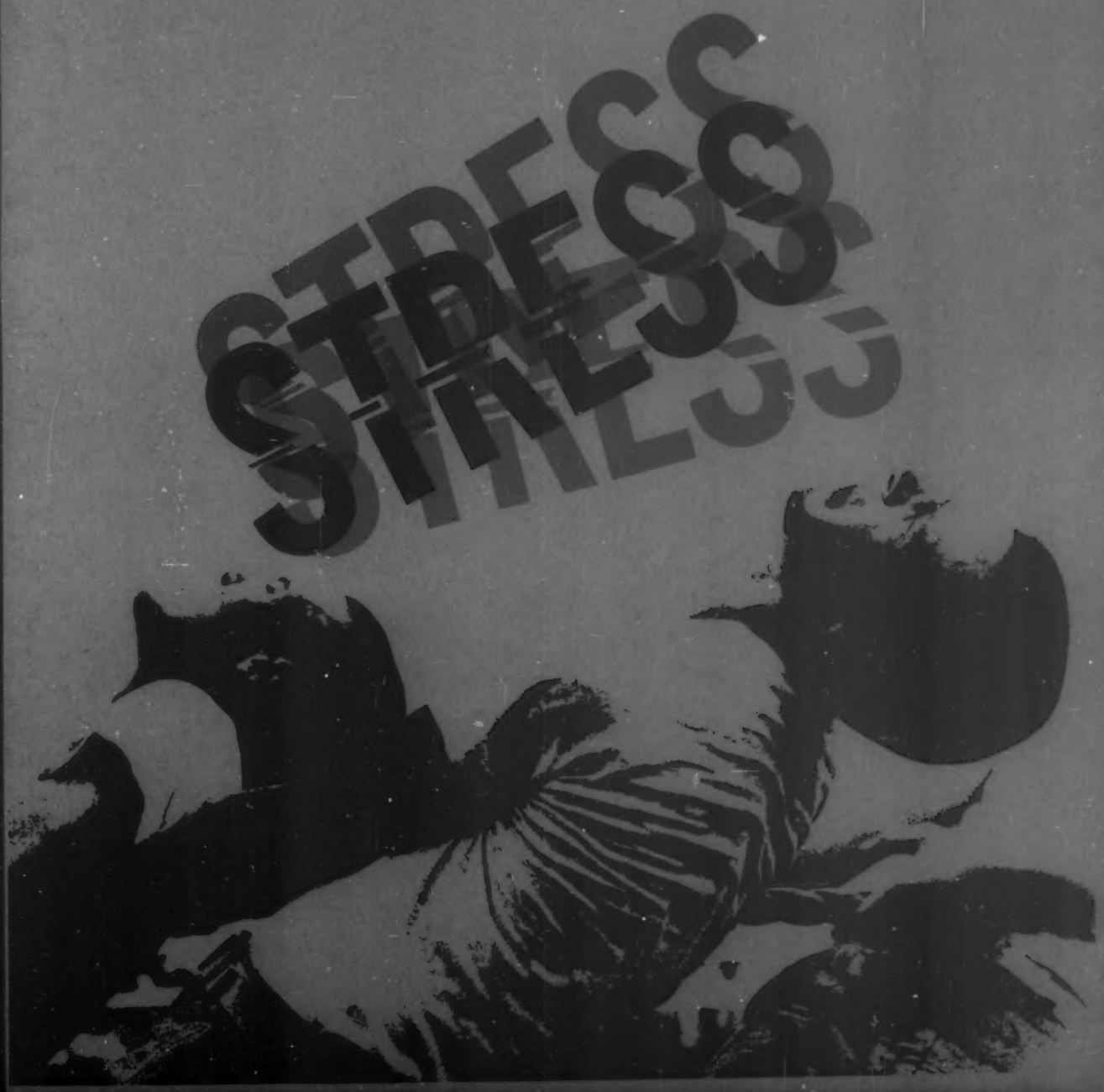

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About Our Cover

The cover picture of Marine recruits engaged in tug of war depicts a type of physical stress experienced during basic training. Physical and mental stresses are an ever present part of military life. The article beginning on page 2 describes research pertaining to the psycho-neuro control mechanisms involved in responses to stressful situations.

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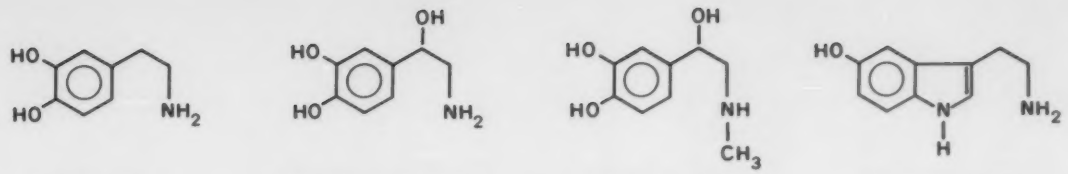
Neuroregulators And Stress

by Jack D. Barchas, Ivan N. Mefford, Kevin Roth, John Madden IV, and Patricia R. Barchas
Stanford University

Acute and chronic stress situations are an ever present part of living, especially in certain military situations. As yet our ability to minimize the degrading effects of too much stress or, contrary wise, to take full advantage of appropriate stress levels is limited. In large part, the limitations exist because of our lack of knowledge regarding the complex physiological interactions among a number of neurotransmitters, neuromodulators, and hormones, to mention only a few of the many chemicals of the body.

Operational conditions expose naval personnel both to environmental as well as emotional stresses. Most Navy recruits, for instance, are exposed in basic training to an alien life style because of the strict discipline and extreme physical challenges. Well established Navy problems causing stress are long working hours with disturbed sleep patterns, exposure to life and death situations, and long-term separation from families. These conditions can lead to decreased efficiency and attrition of personnel. Understanding the psycho-neuro control mechanisms involved in the responses to such experiences is basic to alleviating or, at least, decreasing, their effects. The research described in this article by Dr. Barchas is aimed at delineating the role of various transmitters and hormones as they might be involved in the stress response.



DOPAMINE | NOREPINEPHRINE (Noradenaline) | EPINEPHRINE (Adrenaline) | SEROTONIN

Figure 1. *Chemical structures of some of the biogenic amine neurotransmitters and hormones under study.*

Introduction

The Navy is concerned about stress situations encountered by its personnel and how they handle them. The effects of stress at a biochemical and psychological level are important in terms of the success of the individual in coping and surviving. A striking development in the basic science of stress has occurred over the past two decades. That development deals with chemicals that transmit information, either as hormones in the periphery of the brain or as neurotransmitters in the brain.

Scientists at the Pritzker Laboratory, Stanford University have been conducting a comprehensive research program on the biochemistry of stress; this program, which is funded by the Office of Naval Research is essentially a five-pronged program dealing with: 1) detection of new stress related compounds and study of their biochemical regulation, 2) development of improved analytical methods for stress-related compounds using high performance liquid chromatography and mass spectrometry, 3) localization of stress hormones in brain and peripheral tissue using histochemistry with light and electron microscopy, 4) investigation of the effects of stress using animal models, and 5) studies in humans using Naval and civilian populations. Each of the sections below involves a combination of these approaches. They are a sampling of studies from which we are gathering data on peripheral and central nervous system catecholamines and endogenous peptides.

Peripheral Catecholamines and Stress

A series of experiments examined the relationship of plasma hormones (epinephrine and norepinephrine, shown in Figure 1) to a variety of stresses in humans. Norepinephrine in blood plasma is derived primarily from the nerve fibers in the sympathetic nervous system going to various organs and blood vessels. Plasma epinephrine (adrenaline) comes primarily from the adrenal medulla, an organ which has as one of its major functions the formation and secretion of epinephrine.

Since the early 1950's, the measurement of the amounts of these compounds in urine in response to stress or a psychological state has been used to relate these hormones (catecholamines) to different types of stress. Epinephrine in urine is increased during states of anxiety, anticipation, lack of control, or during mental work. Norepinephrine in urine is increased after involvement in angry or aggressive displays as well as after physical activity.

There are many difficulties with measurement of catecholamines in urine and the amounts present in blood have been sufficiently low that analytical problems prevented accurate assay. These difficulties persisted for a number of years despite the fact the epinephrine was the first hormone crystallized, an event which occurred at the turn of the century. Our intent has been to make measurements of plasma catecholamines on a continuous basis while psychological and physical stressors are applied to the subjects, thus observing resting levels as well as the magnitude and time course of the hormonal responses to these stressors. This has required the development of suitable devices to allow continuous collection of blood plasma. It has also required a method of analysis that would facilitate measurement of large numbers of samples containing extremely small quantities of these compounds. Both of these goals have now been accomplished and the results are now being applied.

The apparatus for continuous collection of blood was designed to allow the subjects some freedom of movement throughout the testing procedure. One end of a small polyethylene tube, treated to prevent clotting and sterilized, is placed in the forearm vein of the subject. The other end is connected to a minia-

Jack D. Barchas

Ivan N. Mefford

Kevin Roth

John Madden

Pat R. Barchas

Jack D. Barchas, M.D., is the holder of the Nancy Friend Pritzker Professorship in Psychiatry and Behavioral Sciences at the Stanford University School of Medicine. Dr. Barchas is also Director of the Nancy Pritzker Laboratory of Behavioral Neurochemistry at Stanford. A member of the Institute of Medicine of the National Academy of Sciences and recipient of a number of awards, he gave the Suzanne Kronheim Lecture at the 1981 meeting of the Undersea Medical Society.

Dr. Ivan N. Mefford is a Research Associate at Stanford in Psychiatry and Behavioral Sciences. Dr. Mefford took his doctoral training in analytical chemistry prior to coming to Stanford. He established new analytical techniques and applied those methods to the measurement of hormones and chemicals that are present in the brain in relation to stress.

Kevin Roth is a graduate student in the Medical Scientist Training Program leading to a combined M.D./Ph.D. at Stanford. The focus of his research has been on the neurochemical changes associated with stress.

Dr. John Madden IV is a Research Associate at Stanford in Psychiatry and Behavioral Sciences. He is particularly interested in the biochemical aspects of stress as they relate to psychological processes.

Dr. Patricia R. Barchas is Assistant Professor of Sociology and (by courtesy) Psychiatry and Behavior Sciences at Stanford and is also Director of the Program in Sociophysiology of the Laboratory of Social Research at the University. She has been particularly interested in the ways in which social environmental processes, including social stress, alter the physiology of the individual.

ture pump which draws 1.6 milliliters of blood every two minutes. The blood is placed in small tubes and mixed with an anticoagulant by a microfractionator which fills one tube every two minutes and automatically moves to the next tube. Red cells are separated from blood plasma which is then frozen until analysis. When all plasma samples have been collected, analysis for catecholamines takes place.

Development of these procedures involved development of a technique capable of measuring as little as 10 picograms (10^{-12} grams) of epinephrine per milliliter of plasma. The method chosen for this analysis was high—performance liquid chromatography with electrochemical detection, a technique made practical be researchers at the University of Kansas under direction from Dr. Ralph Adams and which we have further refined. This technique involves isolation of the catecholamines on alumina followed by separation of the individual compounds on a chromatography column. The chromatography column consists of fine silica beads (5 μ in diameter) coated with a monolayer of long chain hydrocarbon. These beads are packed under high pressure into a 25 cm stainless steel tube.

To accomplish the separation on this column, the sample is introduced into the top of the column and carried through it by a continuous flow of solvent (water, methanol, etc.). As various compounds pass by the coated silica particles, they are attracted to the hydrocarbon coating. The degree to which they are attracted and thereby delayed in passing through depends on their properties. In this way, individual compounds may be separated. The sensitivity to detect picogram quantities is obtained by using an electrochemical technique to detect the compounds as the solvent carries them off the column.

The compounds of interest, norepinephrine, and epinephrine are detected by an electrochemical process, oxidation. A voltage difference is maintained between

a reference electrode (a silver wire coated with silver chloride) and the detector electrode of carbon paste (high purity, finely divided graphite mixed with an inert mineral oil). A simplified circuit diagram is shown in Figure 2.

As the catecholamines pass the detector electrode, the voltage difference between the reference and detecting electrodes causes the molecule to give up electrons. When this occurs, Faradaic current flows through the system and can be detected. Because the detector electrode is in a flowing stream at the end of the column, the sensitivity is very good. With great care in preparation of the electrodes and solvents, the requisite picogram sensitivity may be obtained. Additionally, the cost of the system is relatively small, and it is durable and relatively simple to operate.

By coupling the two techniques described, it has been possible to assess human subjects in the laboratory. The data show that norepinephrine increases markedly after venipuncture, knee bends, mental arithmetic, and arm immersion in cold water. Maximum levels are usually achieved within 2 minutes of the initiation of these interventions. The duration of the elevated norepinephrine response is somewhat longer following cold pressor than for the other physiological stressors. The norepinephrine system responds rapidly but also returns to baseline quickly, with resting levels achieved after a 15-minute rest interval in most cases. The epinephrine response was quite marked during mental arithmetic and also after the cold pressor task. The duration of the response for epinephrine was even shorter than that seen for norepinephrine.

The use of continuous-sampling techniques will enable study of the kinetic patterns of catecholamine release in humans, an area that has previously been impossible for this major stress-hormone system. Since earlier studies have shown the half-life of norepinephrine in plasma to be about 2.5 minutes and epinephrine to be approximately 1.2 minutes, the continuous-sampling technique may allow observations not previously seen with intermittent sampling. In studies examining metabolites that cannot be examined by the assay procedures described above, gas-chromatography and mass-spectrometry procedures are being utilized to measure concentrations of substances and identify new metabolites.

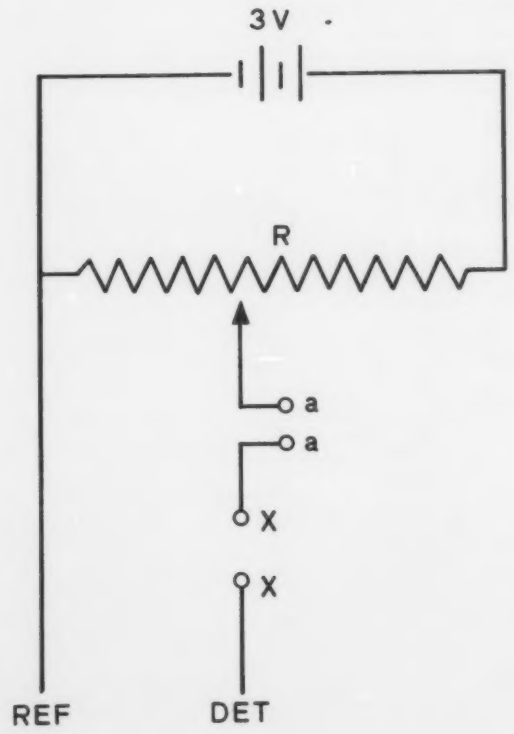


Figure 2. *Circuit design for electrochemical detector used in analytical procedures. The detector is coupled to a high pressure liquid chromatography system and utilizes a reference and detector electrode. R - resistor; a, a - current measuring device; x,x - current offset connections.*

Brain Catecholamines and Stress

Nerve cells within the brain act through both electrical and chemical means. Information is transmitted along a nerve cell by electrical conductance, but at the crucial junction where one nerve cell meets another (the synapse) the process of communication is chemical. A chemical compound released from one nerve cell to another either stimulates or inhibits the possibility of an electrical impulse in the second neuron. Some chemicals are released from one nerve cell to another in a very direct manner and are referred to as neurotransmitters. Other chemicals, referred to as neuromodulators, may be released at one site and influence nerve cells some distance away or may alter properties of the second neuron in a different manner. Chemicals that act as either neurotransmitters or as neuromodulators are referred to as neuroregulators.

Some of the processes involved in neurotransmission are formation of the transmitter, its storage in the nerve cell, its release to the second neuron, and its metabolic inactivation. A schematic of these processes

is presented in Figure 3 which demonstrates aspects of neurotransmission involved in nerve cells that use the neurotransmitter dopamine in communication.

There are several catecholamine neurotransmitters just as there are several catecholamines that function as hormones in the periphery. The two pools are essentially separate. Dopamine is well known for its role in movement but also has roles in behavior. Norepinephrine is important in attention, sleep, and learning. These two materials, dopamine and norepinephrine, are widely distributed in the brain as shown in Figure 4 which also shows the mapping in the brain of still another important neurotransmitter that changes dramatically in stress, serotonin. It has been shown in rats that cold water swimming will dramatically decrease brain norepinephrine and increase brain serotonin. Thus, these two important transmitter systems behave differently. An understanding of the mechanisms by which changes occur could be important to altering or improving the response to stress.

Change in the Rate-Limiting Step of Catecholamine Formation in Stress

One of the most striking aspects of the changes associated with stress occurs at a basic biochemical step in the formation of catecholamines. As shown in Figure 3, the first-step in the formation of catecholamines is the conversion of an amino acid found in the diet, tyrosine, to dopa. Dopa proceeds through a series of steps to the transmitters dopamine, norepinephrine, or epinephrine in nerve cells. The first step, conversion of tyrosine to dopa, is performed by the enzyme tyrosine hydroxylase and is rate-limiting in that it controls the entire pathway. In times of stress the activity of the enzyme is markedly changed and changes of up to 10-fold are possible.

The importance of the enzyme to the stress response made it an attractive candidate for investigation by the Pritzker Laboratory. Although it was discovered over 15 years ago, the enzyme had proved resistant to study. After intensive efforts the enzyme was purified from brain matter and its behavior characterized. The addition of a phosphate group to the enzyme markedly changes its rate of activity. Conditions such as those involved with stress result in the phosphorylation of the enzyme with increased activity and an increased ability to form dopamine and other catecholamines. Evidence has also been found of a mechanism for removing the phosphate group. There is a range of processes which allows the enzyme to become activated and deactivated in relation to stress.

Earlier studies in this sequence established that the normal resting activity of the enzyme is controlled genetically. It is not yet known whether there are genetic influences on the ability of the enzyme to be activated or deactivated in times of stress. Under

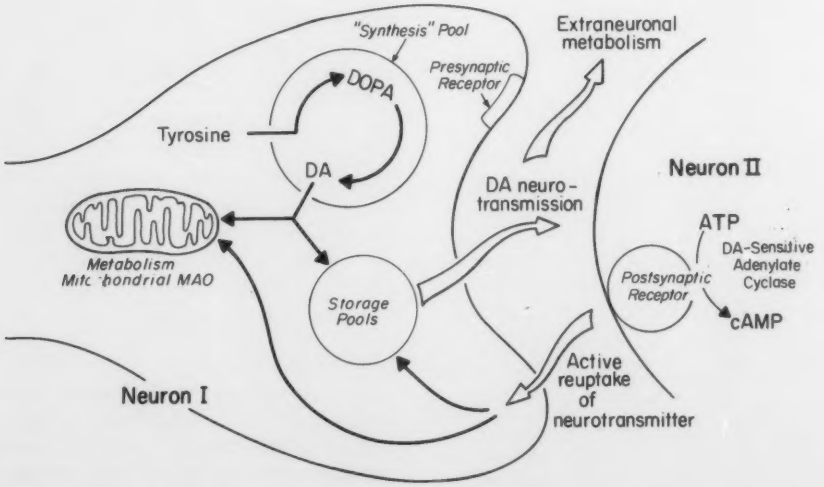


Figure 3. *Schematic of the junction between two nerve cells in which dopamine is a transmitter.*

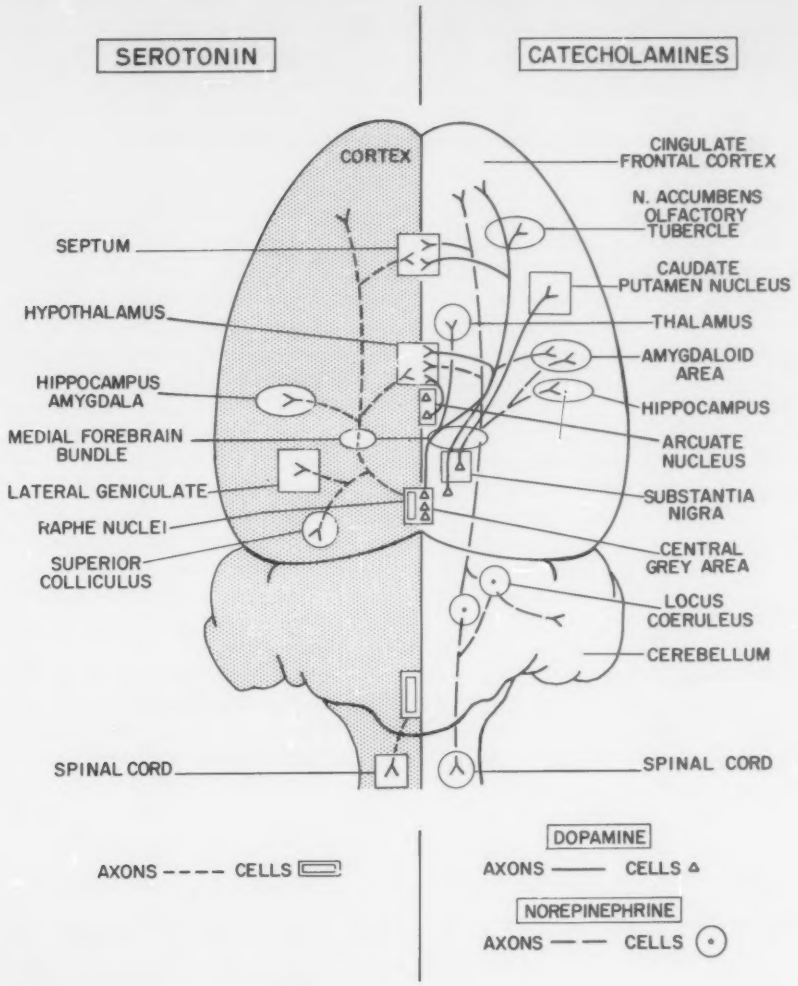


Figure 4. *Diagram of some of the major pathways involving biogenic amines in the brain. Research demonstrating these pathways has come from a number of investigators. Diagram by Dr. Stanley Watson, coauthor in Barchas, et al, Science, 1978.*

stressful conditions some individuals may have variations in this system that prevent successful adaptation and coping with either a relative excess or deficiency or an alteration in the timing of the processes. With reference to some of these possibilities, it has been possible to develop a mathematical model, which is now being tested, of the kinetics of the enzyme.

Changes in Brain Catecholamine Concentration in Stress

The problem of brain catecholamines and stress has also been approached by direct investigation of the changes in the substances with stress. Pritzker Laboratory work has demonstrated that epinephrine could be formed in brain and demonstrated its metabolic pathways. Although brain epinephrine represents only 1-2 percent of the total brain catecholamine concentration, it may control the release of several hormones and modulate the activity of other neurotransmitters. The effects of stress on brain epinephrine are being compared to brain dopamine and norepinephrine in a series of studies.

One study deals with cold water stress. In that

situation, rats were exposed to a brief cold swim (3 minutes in 4 °C water) and the concentration of brain catecholamines examined. Brain epinephrine is extremely responsive to stress and the effects of stress are long lasting. As shown in Figure 5, dopamine concentrations were unaffected by the cold swim, norepinephrine concentrations decreased about 20 percent, and even more dramatically, epinephrine concentrations decreased 30-40 percent. When the effects of cold swim were examined over time, it was found that norepinephrine concentrations recovered within 14 hours but epinephrine concentrations were still decreased 24-48 hours after the three-minute stress.

Summarizing a set of studies, we can say that stress, either acute or chronic, produces dramatic changes in brain epinephrine and in norepinephrine. For example, in one chronic stress study in which various stresses, such as increased housing density, light/dark cycle reversal, and cold swim, were applied intermittently over three weeks, epinephrine turnover was found to be decreased. This altered catecholamine activity both during and after stress may have behavioral consequences.

Opioid Peptide Systems and Stress

An exciting development in relation to neuroregulators was the finding five years ago that the body produces morphine-like (opioid) substances. These materials are found in the brain and the pituitary gland as well as in other areas. Made up of chains of amino acids, peptides, the compounds as a group are termed endorphins. Within that group are a number of subcategories that have led to a confusing nomenclature but a potential richness of function in relation to stress and behavior (see Figure 6 for relations between some of the opioid peptides). The first of the opioid peptides found were the enkephalins, consisting of a chain of five amino acids, now recognized to be widely present in the brain. The brain also contains a larger group of substances of which the best studied is β-endorphin. The β-endorphin and enkephalin systems are quite separate as shown in Figure 7. β-endorphin tracks tend to be long and a number of important areas are involved, while enkephalin-containing cells are present in a large number of areas with shorter connecting nerve cells. Very recently still another powerful morphine-like substance, dynorphin was found in the pituitary gland and the brain. It is now recognized that the inner section of the adrenal gland, the medulla, which normally forms epinephrine, is very active in the production of enkephalin-like materials.

Efforts in the Pritzker Laboratory have focused on three efforts dealing with these materials: 1) identification of new forms of peptides that could be related to stress and behavior, 2) alterations in brain endorphins in relation to stress and behavior, and 3) investigation of the types of opioid peptides present in human blood and determination of the changes that occur with stress.

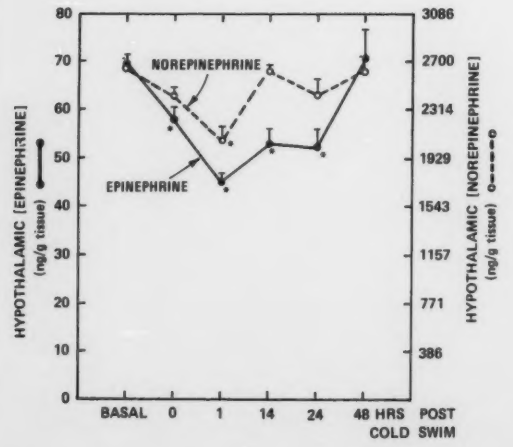


Figure 5. Changes in brain epinephrine and norepinephrine during a 48-hour period after a three minute cold water swim stress.

Identification of New Peptides

In the first area, we have demonstrated several previously unrecognized forms of endorphin present in the pituitary gland (a major gland in relation to stress). Some of these forms have morphine-like actions while others do not, raising the possibility of hormonal endorphin systems that function in quite different ways. The structure of those new peptides is being actively investigated and has been determined for two of them. Studies involving these peptides in relation to stress are now underway.

Alterations in Brain Endorphins in Relation to Stress

The first evidence of a physiologically induced change in opioid peptides came from investigations in the Pritzker Laboratory. It was demonstrated that

OPIOID AND RELATED PEPTIDES

Methionine-Enkephalin (Met-enkephalin)	Tyr-Gly-Gly-Phe-Met (β-LPH 61-65)
Leucine-Enkephalin (Leu-enkephalin)	Tyr-Gly-Gly-Phe-Leu
β-Endorphin (β-END)	β-LPH 61-91
β-Lipotropin (β-LPH)	91 Amino Acids—Contains β-END
Adrenocorticotropin (ACTH)	39 Amino Acids
α-Melanocyte Stimulating Hormone (α-MSH)	Ac-Ser-Tyr-Ser-Met-Gly-His-Phe-Arg-Trp-Gly-Lys-Pro-Val-NH Cleaved from ACTH and Processed Further
Corticotropin-Like Intermediate Lobe Peptide	Cleaved from ACTH
Dynorphin	Tyr-Gly-Gly-Phe-Leu-Arg-Arg-Ile-Arg-Pro-Lys-Leu-Lys-Trp-Asp-Asn-Gln (OH)

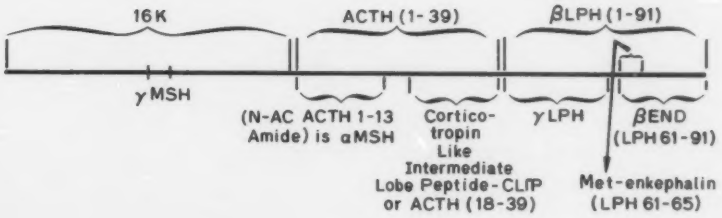


Figure 6. Neuropeptides under investigation in the Pritzker Laboratory. The relationships of some of these related peptides are shown by their common origin from a large precursor petide. The enkephalin and dynorphin systems may have other precursor peptides. Adapted from Watson, et al 1981.

increased levels of opioid peptides in rat brain after stress occurred with concomitant and parallel changes in pain threshold. Recent clinical experimentation by others indicates a similar phenomenon may occur in man following inescapable noxious stimuli. Collectively, these findings may have relevance to human anecdotal reports of changes in pain threshold accompanying certain stresses, particularly those found in a military environment. The psychological dimension of the earlier observations is being extended. A particularly interesting issue is that control over the aversive event may be more important to the physiological stress response than is the presence of the aversive event.

After inescapable and uncontrollable stress, but not after escapable stress, there is a long-term analgesic response that can be activated and demonstrated at 24 hours after the cessation of the stress.

This phenomenon can be prevented by agents which block opioid peptides (e.g. naltrexone) if they are administered prior to the inescapable shock or prior to analgesic testing. This pharmacological action occurs in a dose-dependent manner and suggests a role of endogenous opioid peptides in certain forms of pain-relief mechanisms.

Powerful interrelationships are now being found in this series of studies between stress-induced opioid processes and the pituitary adrenal steroid systems. Specifically, in a collaborative effort with Dr. Steven Maier of the University of Colorado, it has been demonstrated that the integrity of the pituitary-adrenal axis is necessary for the opioid-mediated stress-induced analgesia and that corticosterone (an adrenal steroid) is necessary for the expression of the long-term analgesic response. Another direction for further research involves the relationship between controllability and lack of controllability on endogenous peptides and the immune system, with reference to the organisms' susceptibility and resistance to disease.

Opioid Peptides in Human Blood

The study of opioid peptides in human blood is a particularly interesting research area in relation to stress. If morphine-like materials are being secreted, then it is important to determine the patterns of secretion and the conditions of release. While there are certain difficulties in working with blood, it has been possible to demonstrate opioid-like activity in plasma. Studies in the Pritzker Laboratory have found evidence for the existence of opioid peptides of several different molecular weights. Large numbers of proteins and peptides are also found which generate opioid activity following exposure to certain enzymes. These compounds are different from, and present in much greater quantities than, previously described opioid peptides in human plasma.

Several of the materials present in plasma resemble enkephalins of a relatively large size. The physiological role of some of the proteins and peptides that have enkephalin-like activity, either in their native form or after enzymatic action, is a central question. They may be found to be hormonal and their potential role on peripheral organs or on the brain will be of considerable interest.

In addition to the enkephalin-like substances, there are three peptides of different sizes which resemble dynorphins. It is possible that the blood may contain β-endorphin-like substances from the pituitary, dynorphin-like materials from the pituitary, and enkephalin-like substances from the adrenal gland. Each of these is a potentially stress related system.

Circulating epinephrine, the level of which is doubled following 2 minutes of knee bends, has its origin in cells in the adrenal medulla which are known from the work of various laboratories to contain opiate-like materials. Further investigations of human blood in the Pritzker Laboratory reveal a greater than 2-fold increase in the levels of circulating opioid receptor active peptides with stress, under conditions which also cause a change of similar magnitude in catecholamines. This finding of parallel behavior in an opioid peptide system and a catecholamine system in humans suggests that the hormonal opioid peptide system reflected in blood may be of interest in relation to stress. The *pattern* of response involving the various stress hormones is an area now requiring investigation.

Examples of Studies of the Stress Hormones in Human Populations

Two studies on the responses of young people to stress have been undertaken. The first deals with psychological and physiological responses to military basic training; the second is on aspects of stress in college students in the same age range.

In a collaborative study with the San Diego Naval Facility, under the immediate direction of Dr. Ross Vickers, the changes that occur through Marine basic training are under investigation. For those studies, 96 subjects from four platoons composed the test population. Of those individuals who gave informed consent to be in the study, repeated physiological measures and behavioral responses were taken. This important collaboration with the Navy involves assessment of various catecholamine and opioid and peptide hormonal systems in relation to performance through training, as well as self-reports of mental state and other factors in the social environment including concurrently obtained perceptions of leadership, social influence, health, and platoon environment. It is planned to extend this study to cold-environment training.

The second study deals with the relation between social environment, psychological state, and the same physiological measures in a group of 300 men and women in college, taken as age controls for the men in service. The male and female student volunteers were recruited for blood drives at the college and also consented to be in the study. For this study, they provided responses to the Holmes-Rahe life change ques-

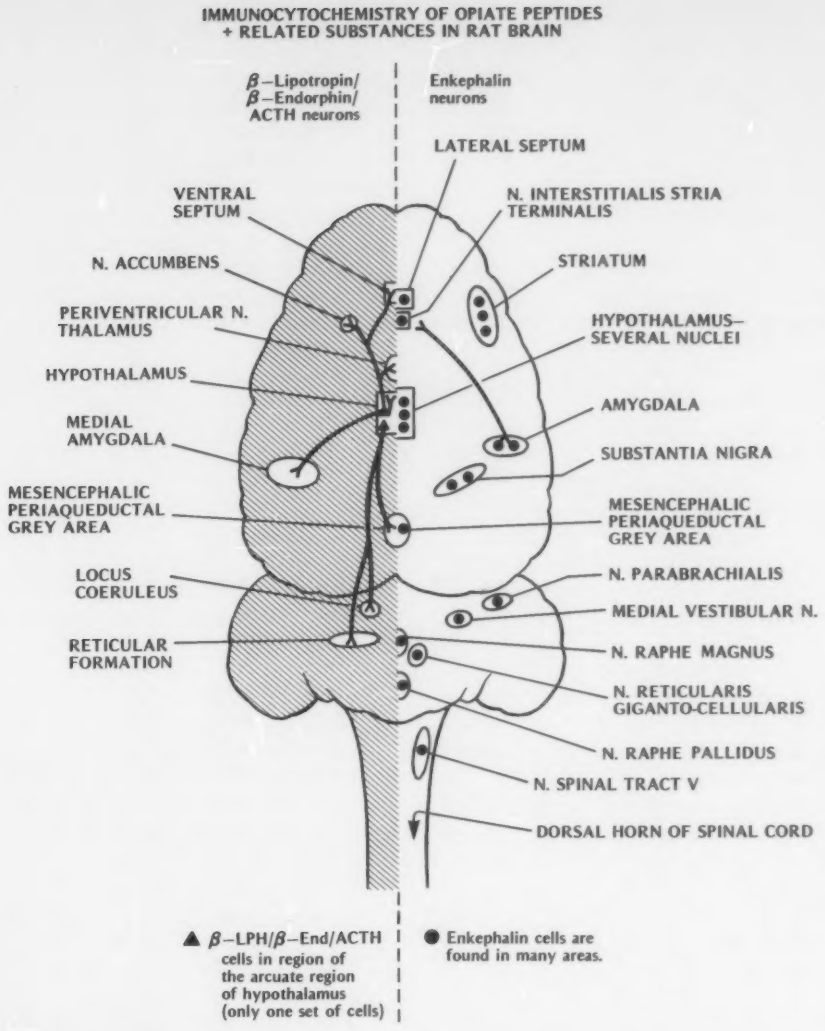


Figure 7. *Diagram of some of the major pathways involving enkephalin and endorphin peptides in the brain. Research demonstrating these pathways has come from a number of investigators. Diagram by Dr. Stanley Watson, coauthor in Barchas, et al, Science 1978.*

tionnaires: its subscales reflect changes in commitment, in personal habits, in personal concept, and in social relationships or the individuals' social nexus, as well as changes in intimate relationships. Type A personality and anxiety measures were also taken.

Thus far, there are marked sex differences when the questionnaire responses are correlated with hormonal responses. For example, the women, but not the men, varied in adrenaline, endorphins and cortisol by dormitory—suggesting that on the whole, housing and immediate surroundings may be more important for women than for men.

For men, however, but not for women, Type-A personality seems to be positively related to both epinephrine and to β-endorphin. In addition, for the men the Holmes-Rahe total life-change questionnaire responses were highly associated with values of norepinephrine, epinephrine and β-endorphin-like materials. (The more life events or changes reported over the last 12 months, the higher the values of the

compounds.)

These studies on stress in young people provide a normative data base for further consideration of stress and coping, and the hormonal reactions that accompany the processes.

Conclusion

Stress can take many forms and, dependent on the type of stress, can impact on a number of different biochemical systems, thereby markedly modifying the behavior of the organism. A more complete understanding of the biochemistry of stress in terms of neuroregulators and hormones should permit earlier recognition of certain types of stress states and improved assessment of stressful environments. Such knowledge should lead to minimization of debilitating stress situations as well as to improved treatment for unavoidable stress-related conditions. Continued basic investigation is required, including elucidation of stress-related mechanisms and application of basic science in the clinical laboratory and natural settings. ■

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profiles in science

Professor Douglas L. Inman, who has been a long time contractor of the Office of Naval Research, is Director of the Center for Coastal Studies, Scripps Institution of Oceanography, University of California, La Jolla, California. During thirty years of research and teaching at Scripps, Dr. Inman has pioneered the field of beach and nearshore processes. Among the first to apply quantitative physics to coastal phenomena, he has helped lead the way to unraveling the mechanics of wave, current, and sediment interactions which mold the shorelines of the world. His conceptual contributions have produced the vocabulary of coastal oceanography: nearshore circulation, littoral cells, reflective and dissipative beaches, collision and trailing edge coasts, and a morphotectonic classification of beaches and coasts. These achievements are now finding applications in coastal planning, harbor engineering, and marine archaeology.

Professor Inman was a member of the American Oceanography Delegation to the People's Republic of China in 1978 and an author of *Oceanography in China,* a guideline for U.S. and Chinese interaction in the field of Oceanography. Coupling an interest in history with his knowledge of shore processes has resulted in studies of ancient harbors dating from Minoan, Phoenician, Greek, Roman, and Crusader periods. He was a Guggenheim Fellow, 1961-62 and is a Fellow of the Geological Society of America. ■

Assessing the Present and Anticipating the Future: A Method For Analyzing World Politics

by

Frederick A. Rothe

International Public Policy Research Corporation

This paper discusses techniques which are employed to anticipate international crises or conflicts. These techniques classify international political activity into one of five classes and make projections about the future condition of the political activity. The techniques are designed to aid the people who are responsible for making current assessments and also future projections, or monitoring and warning. The techniques are demonstrated in the context of the 1980 Iran-Iraq War and other recent world political history. The main purpose of the techniques is to anticipate international crises or conflicts.

Introduction

The Early Warning and Monitoring System (EWAMS) is a computerized system designed to monitor the international political relations among or between all the nations and other important international groups or organizations in the world.[1] The EWAMS is able to track the affairs of a single nation, a pair of nations, a group of nations, or the general political condition of the world. This paper discusses and demonstrates the techniques used to monitor and project the condition of world politics. The technique is called the Systemic Scan Indicator (SSI) and it is the component of the EWAMS that is designed to scan the entire international political system.

The paper is divided into three major sections. The first section offers a discussion about how the SSI actually works. The second section provides the results of the SSI research to date and includes the most intriguing set of findings. The final section discusses how the methods may be used for other research and analysis, but not at the systemic level. Listed below are some of the major findings from the second section of the report.

- The international system is "normal" or "routine" approximately 81 percent of the time and "nonroutine" or "dangerous" approximately 19 percent of the time;
- The international system changes from week to week in an orderly but probabilistic fashion;
- Statistical tests (Chi-square) support the proposition that the current state of the system is the best predictor of the next or future state;
- The statistical tests and the change probabilities indicate that the international system can be treated as a first-order Markov chain.

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[1] *The EWAMS is continuing to be developed and enhanced by the International Public Policy Research Corporation and is sponsored by DARPA/SSD. Many technical reports and research memoranda outline the various components of the EWAMS. Further explanation on the concepts, methods, and data of the EWAMS can be found in the publicly available reports and papers. An overview of the EWAMS project was published in the Spring, 1980, issue of* Naval Research Reviews.

Dr. Rothe is Vice President at the International Public Policy Research Corporation, McLean, Virginia. Dr. Rothe has developed new strategies and quantitative indicators of monitoring and projecting international political affairs. He is the author of numerous papers on international systems analysis, foreign policy studies, and national security affairs.

The Systematic Scan Indicator (SSI)

To reiterate, the Systemic Scan Indicator (SSI) is an indicator of the condition or state of the entire international system as defined by the EWAMS databases. It is a weekly indicator that classifies all international activity into one of five classes. Each of the five classes contains varying amounts of international activity. Nations are not included in the classification; only the actions of nations are classified into the five class typology. Although each of the classes is simply a category (or nominal data), the classes range from very normal or routine (Class 1) to extremely volatile or dangerous (Class 5). Classes 1 and 2 reflect the routine nature of international politics and Classes 4 and 5 reflect the dangerous, conflict-laden aspect of world affairs. Class 3 tends to be a mix of the other four, reflecting both the routine and danger components of world politics.[2] Figure 1 gives a brief narrative summary and examples of each of the classes.

The classification scheme was developed on a sample of 303 weeks surrounding known international crises (Ben-Bassat et al., 1980). Since then, each week of the entire time period from 1966 to the end of 1980 has been classified according to the original 303 weeks, or "training patterns." The 303 classified weeks are used to "train" the "classifier" in classifying every additional week. The "classifier"

[2] *For a more complete discussion of the classification scheme and the methodology used to develop it, see Ben-Bassat et al. (1980) or Rothe (1979).*

Figure 1
Classes of International Political Activity

Classes	Description	Example
1. Normal	Includes most time periods and represents day-to-day diplomatic activity.	
2. Normal with localized incidents	Primarily normal but may include small, localized incidents.	1975 Mayaquez
3. Localized crisis	A blend of normal (Classes 1 and 2) and violent (Classes 4 and 5) activity and may include localized crises; tends to precede the violent classes.	1968 Czechoslovakia
4. International crisis	Violence and the use of force are dominant; considerable global involvement.	1967 June War 1970 PLO-Jordan Confrontation 1971 Laos Offensive
5. International crisis with severe violence	Acute and heavy violence of greater magnitude than Class 4; often the "peak" weeks of a crisis and includes much global activity.	1973 October War 1971 India-Pakistan War 1970 Cambodian Incursion

now in use at the International Public Policy Research Corporation (IPPRC) is a form of discriminant analysis known as "nearest neighbor" discriminant analysis (Patrick, 1972). The nearest neighbor classifier works in the following fashion.[3]

Data for each week are retrieved from the EWAMS and are appended to the 303 training patterns. The format of the data is the date (beginning date of each week), the values for each of the 22 major events, and a classification (1 through 5) of the week. The analyst adds the classification of the week, but this information is for "data management" purposes and does not have an impact on the classification routine. Thus, any of the five classes can be added on to the current week's data.

Once the data have been organized in this fashion, the nearest neighbor classifier is used to classify the weeks that have been appended to the original 303 training patterns. The significant difference between nearest neighbor discriminant analysis and classical discriminant analysis is that the nearest neighbor does not require linear separation between classes (Cooley and Lohnes, 1971; Patrick, 1972). The nearest neighbor simply takes the new week and finds the week of the training patterns to which it is most similar—its nearest neighbor. The new week is then given the classification of the week to which it is most similar or closest. A Chi-square distance measure is used to calculate this distance. A Chi-square measure is used not only because it accounts for the values in each of the 22 event categories, but also because it accounts for the total number of events in each time period. Total volume of events is one of the best measures for determining the condition of the international system or the relationship between two countries.

The nearest neighbor classifier at IPPRC has one additional option. It is that the analyst can instruct the computer program to classify the new week to more than one neighbor. The single neighbor classification routine is known as the classical nearest neighbor, but it is possible also to get a majority of neighbors. In the example that follows, a total of three (3) neighbors was selected. This means that the program selects the three nearest neighbors and then uses a majority rule for classifying the new week.

[3] *In the following discussion, it is important to remember that none of the retrieval, classification, and other analytical routines is currently in the EWAMS. The different components of the SSI are in a "stand-alone" format until they have been thoroughly examined.*

```
Type the number of neighbors you would like  3
Type the number of the pattern at which you want testing to start  1
Type the number of the pattern at which you want testing to stop  314
Type the number of training vectors you wish  303
Type the number of the pattern at which you want printing to start  310
```

Table 1
Instructions for the Nearest Neighbor Classifier

Therefore, if the three nearest neighbors are a class 4 and two class 3s, the program will classify the new week as a class 3, even though the single closest neighbor is a class 4.

Table 1 displays the instructions for using the nearest neighbor classifier. The first line permits the analyst to select the desired number of neighbors. Any odd number (because of the majority rule) from one to nine is acceptable. The second and fourth lines always remain the same; testing or classifying must start at the first training pattern and there must be a total of 303 training "vectors" (patterns). The response to line 3 is flexible, depending on how many weeks are to be classified. In our example, we are classifying 11 weeks, so we want the testing to stop at week or pattern 314. The final line is used to control the classifications which are to be printed. In our example, we want to see only the last or most recent five weeks.

Displayed in Table 2 are the results of the classification. The right hand column shows the distances of the new week to the nearest neighbors. The middle column gives the classifications of the three neighbors (from the original 303 training weeks) and the dates of the neighbor weeks are in the left hand column.[4] The five weeks that are classified are the first five weeks of the Iran-Iraq War. If the classical nearest neighbor is invoked, each of the weeks would be a class 4. If the majority rule is invoked, the first week (800914) would be a class 3 and the remainder would be class 4.

One of the benefits of this type of classification is that the analyst can see how the current time period compares with historical time periods. For example, in the first week (pattern 800914), the nearest neighbor is the week ending on 700411. A review of history reminds us that the week of 700411 was in the midst of much violent and conflictual activity occurring two weeks prior to the American incursion into Cambodia. An advantage of displaying the nearest neighbors of the week being classified is that the neighbors offer the analyst a historical perspective on the current time period. Similarities and differences between historical time periods and the current time can be found. This may help the analyst in his/her evaluation and assessment of the current time period.

Table 3 displays the final results of the classification scheme. The values in the right hand column (entitled "classified into cluster") are the results of the classification and are in fact the values of the SSI. The values in the middle column are the classes selected by the analyst at the very beginning of the data retrieval procedure.

These values could be 1s or 2s and are required only for the classification program to work. The results are from the majority rule classification. The single most striking aspect of the analysis is that for the four weeks (800921 to 801012), the international system was in a very dangerous, conflict-laden state. The SSI reflects clearly that during those four weeks the international system was in fact experiencing an international crisis. Affairs between Iran and Iraq were sufficiently violent and far reaching to affect the performance of the entire international system. In this sense the Persian Gulf War was an international crisis.

Values for the SSI are the results of the classification routine. In Table 3, values for the SSI are 3, (800914) and then 4 for the remaining weeks. Even with all the moderately sophisticated statistical manipulation, the final results are values ranging from one

[4]*Because a different data retrieval program was used in the original research and the classification scheme, the dates are the end date of a week, not the beginning date.*

```
pattern 800914:
neighbor        category        distance
  700411            4         0.103e+02
  680831            3         0.120e+02
  760110            3         0.148e+02

pattern 800921:
neighbor        category        distance
  700411            4         0.199e+02
  680831            3         0.227e+02
  730113            4         0.239e+02

pattern 800928:
neighbor        category        distance
  700411            4         0.144e+02
  760110            3         0.184e+02
  700328            4         0.229e+02

pattern 801005:
neighbor        category        distance
  700110            4         0.172e+02
  700328            4         0.186e+02
  700321            4         0.233e+02

pattern 801012:
neighbor        category        distance
  670114            4         0.147e+02
  670715            4         0.189e+02
  700328            4         0.203e+02
```

Table 2
Nearest Neighbor Classification Results
800914–801012

(1) to five (5). A tremendous amount of data is reduced to achieve these values, but this does not mean the data are "lost" or "thrown out." The data are simply grouped together to give a result that is easy to interpret and understand. Although the classification of the time periods is useful in its own right, a more important consideration is to analyze how the international system changes over time. It is important to know the current state (or class) of the international system, but it is equally or more important to know the state to which the system is going to change. By knowing the properties or characteristics of system change, it is possible to forecast (give warning) system states as well as monitor (classify) them.

Change Characteristics of the International System

Monitoring in international affairs deals primarily with recognizing, with some degree of certainty, the current condition or state of relations between two nations, a group of nations, or the entire international system. The SSI satisfies this need; it classifies or recognizes the current condition of the international system. From this perspective the SSI is a good monitoring technique.

Warning in international affairs deals primarily with projecting, again with a degree of probability, what will be the next condition or state of the relations between two nations or the international system. By classifying all the weeks from 1966 to the present and constructing a table of how the SSI changes from week to week for the entire data his-

date	from cluster	classified into cluster
800914	4	3
800921	4	4
800928	4	4
801005	4	4
801012	4	4

The word "cluster" is from the original research. Class, category, system state, or any other word reflecting the different components of a typology can be used in place of cluster.

Table 3
Majority Rule Classification Results

tory, a table of transition probabilities is derived. These transition probabilities are used to project what next week's SSI will be. When the SSI data are organized in this fashion, the SSI is used for warning. A discussion of how this is done will clarify the thought.

All the weeks from 660102 (the first Sunday of 1966) to 801227 have been classified according to the SSI scheme. All of these weeks (a total of 783) are arranged in chronological order in a separate data file. A small program was written to go through this file and tabulate the number of times that each of the five SSI classes followed in time each of the others. The results are displayed in Table 4.

In each cell of Table 4 are two values. The top value is the total number of changes from one class to the others. For example, in the upper left hand cell is the value 176. This means that when the international system was in a class one (C1 for brevity) in one time period, it stayed as a C1 the following time period for 176 out of 321 total time increments. Similarly, reading across the top row in the table, C1 changed to C2 a total of 58 times out of the total of 321. The bottom value in each cell gives the proportion of the change from one class to the others from the total number of changes for that class. Referring to the upper left hand cell again, the .55 means that when the international system is a C1 at one week, 55 percent of the time it will be a C1 the following week. The percentage values form the matrix of transition probabilities, or the probabilities of change from one time period to the next.

At the bottom of Table 4 are the column totals and, below them, another group of percentages labelled "Priors." The "Priors" values are the prior probabilities of a new week being classified into one of the five classes based on the historical data.[5] For example, if we knew nothing of the previous week, we could say that the current week has a .41 probability of being classified into C1. Prior probabilities are used in Bayesian classification procedures (Afifi and Azen, 1972). For the purposes of this paper, the priors are used simply to illustrate the proportion or percentage for each class in nearly fifteen years of recent international political history.

The priors demonstrate that the most extremely conflict laden class (C5) appears very rarely (.01). This class is composed among others, of the peak weeks of the 1971 India-Pakistan War and the 1973 October War in the Middle East. The next most conflict laden class (C4) has appeared more frequently (.18), but is by no means dominant. The most normal or routine classes (C1 or C2) combine for a total of 60 percent of international activity and the remaining class (C3) makes the proportion of more or less normal weeks a total of 81 percent. The international system, therefore, is tranquil and peaceful 81 percent of the time, and is conflict laden about 18 percent.

These findings point to the notion that the international system is really quite stable. To fully understand the concept of stability, the patterns of change, or the transition probabilities, must be analyzed. W. Ross Ashby, one of the original proponents of cybernetics, believes that stability or equilibrium occurs when, for each succeeding time increment, the system stays in its current state or changes to those states to which it is closest or most similar (Ashby, 1956: 73, 77). Table 4 displays this for every state or class. For example, when the international system is in class (or state, the terms

[5] *The grand total number of weeks in Table 4 is 782, or one less than the total number of weeks in the analysis. The difference of one (1) is because the very first week in the analysis had no previous week from which to change.*

Time T-1 Class	Time T Class 1	2	3	4	5	Total
1	176	58	62	25	0	321
	.55	.18	.19	.08	.00	1.00
2	55	60	22	12	1	150
	.37	.40	.15	.08	.01	1.00
3	57	20	47	37	2	163
	.35	.12	.29	.23	.01	1.00
4	32	11	29	61	4	137
	.23	.08	.21	.45	.03	1.00
5	0	1	3	3	4	11
	.00	.09	.27	.27	.36	1.00
Totals	320	150	163	138	11	782
Priors	.41	.19	.21	.18	.01	1.00

Chi Square = 275.57 Degrees of Freedom = 16

Number of time increments is 787.

Table 4
Frequencies and Transition Probabilities of the International System

are used synonymously) C1, it remains in C1 or changes to the next most similar class (C2) 73 percent of the time. Class C2 remains the same or changes to C1 or C3 in 92 percent of the cases. In the more violent classes of the international system, C4 remains the same or changes to its nearest classes (C3 and C5) for 69 percent of all of its changes and C5 changes to C4 or stays as a C5 for 63 percent of all of its changes.

An important aspect of this stability or maintenance of equilibrium is that few surprises occur. It is extremely rare that the international system "jumps" from a normal time period to a dangerous and volatile time period. Not unless the system is in state C3 is there a substantial probability of changing to a C4 state. Also, because of these historical patterns of international system change characteristics, a projection can be made, with a degree of certainty, about what will be the state of the international system in the next time period. In other words, each week a warning can be made, based on the current state of the international system.

This observation brings attention to the single most important value in Table 4—the Chi-square statistic given at the bottom of the table. A Chi-square of 275.57 with 16 degrees of freedom is significant at the .001 level.[6] What this means is that not only is it possible to make probabilistic projections for which class the international system will be in next week, but that those projections can be made

[6] *It is customary not to use a Chi-square test on a matrix which may include an empty or zero cell (Garson, 1971: 134). But the two zero cells in Table 4 are extremely significant from an international politics perspective and must remain in the table. In addition, the Chi-square value is so high that the zero cells make little difference in the final results.*

with confidence. The Chi-square statistic demonstrates that the state of the system at time T-1 is the best predictor for the state of the system at time T.

Although this may appear to be a simplistic and commonsense finding, its significance should not be overlooked. First, this was the major proposition that guided the World Event Interaction Survey (WEIS), of which the Early Warning and Monitoring System (EWAMS) is the descendant. In one of the first, and only, published pieces from the WEIS endeavor, the originator of WEIS and one of his associates stated this proposition only for individual nations:

> We assume that there are continuities and regularities in the international political behaviors of nations and, therefore, that our basic theoretical orientation is tenable: that the past behavior of a given country in the international system is a source for the prediction of its present and future behavior (McClelland and Hoggard, 1969: 712).

The same proposition is applied to the international system; the past behavior of the international system is a source for predicting its present and future behavior. The Chi-square test applied to the SSI matrix is the first time that this proposition has been tested. After nearly fifteen years of effort with a variety of techniques (exponential smoothing, power spectral analysis, Box-Jenkins time series analysis),[7] the original proposition is demonstrated to be correct. In addition, the Chi-square value and the matrix of transition probabilities demonstrate that the international system is a first-order Markov process.

The significance of this basic research finding for the EWAMS is that it is now possible, at the systemic or global level, to give warning. Probabilistic projections about the state of the system in the next time interval can be made. The SSI, therefore, both monitors (by classifying the current week) and warns (by using the matrix of transition probabilities). The warnings are made with confidence that the current week is the best predictor of the next and a probabilistic statement can be made about which of the five system states is the most likely to occur in the next week.

The capabilities of the SSI obviously apply only to the systemic level of monitoring and warning. Intelligence analysts and other potential users of the EWAMS generally are concerned with international regions, a pair of nations, or a single nation. Given the success of the SSI from the basic research perspective and its potential or application, it seems that it would be beneficial to develop a classification scheme for the action of a single nation, pairs of nations (dyads), or regions. How this can be done is discussed next.

Applying the SSI Techniques To Other Levels of Analysis

Applying the SSI methodology to a dyad (pair of countries) or an individual nation, presents no difficulties. The research methodology can be applied to any level of international affairs, and the specifics of the method have already been tested. Thus, much of the time that was spent initially testing the appropriate clustering method can now be saved. The steps for developing a classification scheme for a dyadic relationship are as follows.

First, data for a dyad (e.g., Israel and Egypt) are retrieved around known crises in the country pair's relationship. This is done to capture both the routine aspect of the relationship, as well as the nonroutine or conflict aspect of the relationship. Using Israel and Egypt, it may be desirable to collect all of the events that each nation sent to the other (a "two-way flow" in EWAMS terminology) before, during, and after the 1973 October War, the 1967 June War, and the signing of the Camp David accords. These data are placed in a separate file and form the basis of the classification scheme.

Next, in order to develop the classification scheme, the time increments for the dyadic relationship are clustered together using an agglomerative hierarchical clustering algorithm with a Chi-square distance measure. This algorithm groups together the time periods most similar to each other. The values of each of these 22 event types are the data that the algorithm uses to group together the time periods. In social science terms, the "cases" are grouped together, and the "variables" are the items that determine the similarity of the cases. The outcome of the algorithm is a tree structure with the most similar or closest time periods grouped according to the quality of the dyad's relationship at those points in time.

Clusters are (or should be) apparent in the tree structure. Each of the time periods in each of the clusters is labelled with their cluster or number (1 through 5, for example). The accuracy of the clustering algorithm is then checked using discriminant analysis. Severely misclassified weeks, as evidenced by the results of the discriminant analysis, are relabelled according to their reclassifications, again

[7] *For the single best summary of these efforts, see McClelland et al. (1971).*

determined by the results of the discriminant analysis.

Once these adjustments and corrections of the original clustering scheme are made, the classification scheme is established. The weeks comprising the classification scheme then become the set of "training patterns" used for future classification of new time periods of that dyad. It may be possible to classify other dyadic activity according to the training patterns derived from one particular dyad. Regardless, once the training patterns are established, new weeks in the dyad's relationship can be classified according to the nearest neighbor classifier used in the SSI classification.

It does not matter whether the data "fed into" the clustering and classifying algorithms are from the global, regional, dyadic, or national level. The various algorithms can be used on any level of analysis; thus, the level of analysis question is irrelevant as far as the technical aspects of the procedure are concerned. And the proposition guiding the research—a nation's foreign policy behavior during the present is the best predictor of its future performance—also can be applied to any level of analysis. What cannot be transferred from level to level are the findings of the analysis. It is not correct to infer that, because the international system has five states, a dyad will. A particular dyad may have only three states or it may have seven. The technical aspects of the analysis are consistent across all levels of analysis; the findings may not be.

To conclude this section, it is important to reiterate the rationale for undertaking the classification research. More than anything else, policy practitioners, intelligence analysts, and decision makers of any kind must, before any action is taken or decision is made, correctly identify the *current* state of affairs. Action of any type should be influenced by an accurate identification of the current situation and a fairly certain anticipation of what the situation will become. Whether for the international system as a whole or a single dyad, the classification scheme and its accompanying matrix of transition probabilities meet the requirements of monitoring and warning, or recognition and anticipation. The classification scheme defines with rigor the current state of affairs; the matrix of transition probabilities offers probabilistic anticipation of what the state of affairs will become.

Conclusion

The intent of this paper has been to discuss what the Systemic Scan Indicator (SSI) is and to demonstrate how it works. Emphasis was placed on its capability to both monitor the present state of work politics and to project or provide warning about the next state of world politics. A basic research finding was that there is probabilistic order in the dynamic of global affairs. This finding supported the observation that the trajectory of the international system can be treated as a first order Markov chain, and therefore, probabilistic predictions to the next time interval can be made. The paper then concluded with a discussion about how the SSI algorithms and techniques can be applied to other levels (dyadic, national, or regional) of analysis.

Obviously, the SSI monitors only all of the actions of all nations; the actions of an individual nation or a dyad are not singled out for closer scrutiny. Thus, the specifics of a nation's foreign policy or the interactions of a dyad are not included in the analysis. But, because the international system, and hence the SSI, reflects a local conflict (like the Iran-Iraq conflict mentioned earlier), the SSI can be used as the initial "broad sweep" to find disturbances *anywhere* in the world. More importantly, because of the temporal patterning of the SSI (demonstrated in the matrix of transition probabilities), projections regarding the continuation of a local conflict can be made. World politics are such that localized disturbances tend to dominate the international system. Once the participants of these localized disturbances are known, the SSI is used to make projections about the specific disturbance, because of the tendency of these conflicts to dominate the international system.

International systems analysis and its connection with the demands and requirements of warning and monitoring is still in its formative stages. But enough knowledge has been gained to demonstrate that patterns and orderly change sequences of the international system do exist. This realization alone emphasizes that the affairs of nations are not totally random and chaotic and that, given the appropriate techniques, much of the uncertainty surrounding global politics can be eliminated and predictions can be made. ■

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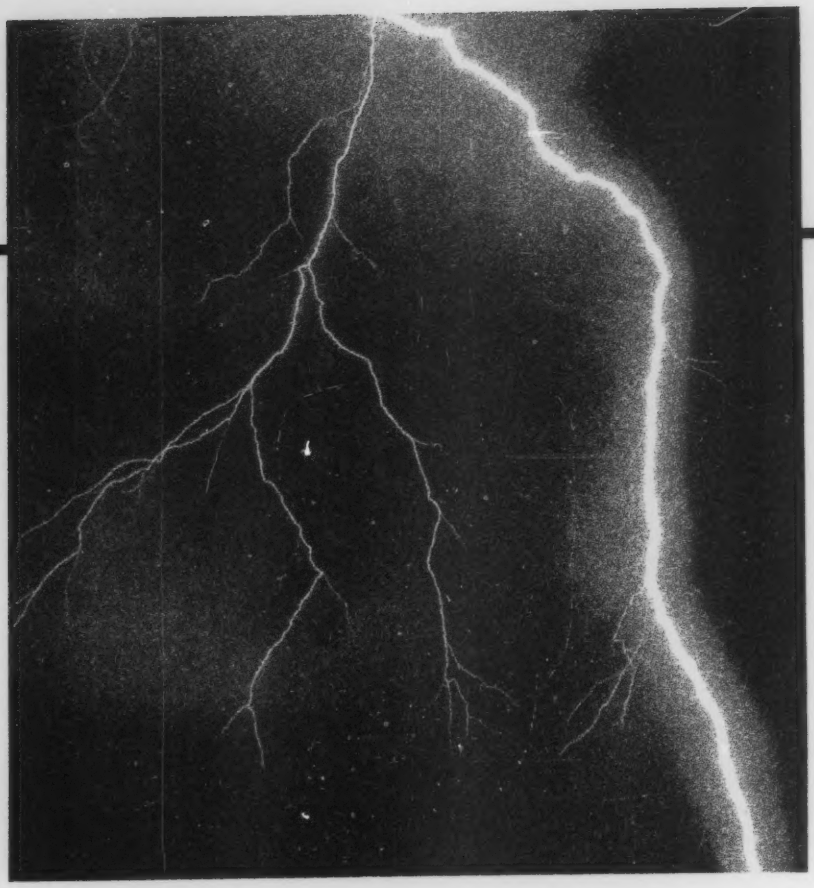
Navy Supports Nobel Laureates

Two of the three Nobel Laureates selected for their work in Physics on October 19 are supported in part by the Navy's Office of Naval Research (ONR) through its Contract Research Program. The two ONR-supported scientists to receive the Nobel Prize this year are Professors Nicholaas Bloembergen of Harvard University and Arthur Schawlow of Stanford University, noted for their pioneering research in the field of laser spectroscopy. The third Nobel winner was Professor Kai Seigbahn of Uppsala University, Sweden, who was honored for his work in electron spectroscopy.

Professor Bloembergen's work, which ONR has helped to support for 30 years, is the culmination of a sustained program of research in the interaction of high intensity laser beams with matter—a discipline known as nonlinear optics. Today, Bloembergen is recognized for developing the technique which focuses three high-intensity laser beams through a substance and produces a fourth beam. This fourth beam can be controlled to produce additional wavelengths of light. His project has the ongoing support of ONR.

Professor Schawlow was also supported by ONR early in his career when he was one of the scientists in the late 1950's responsible for helping to develop the laser from its "parent," the maser. The important technological advance for which Schawlow was awarded the Nobel Prize was the development and application of laser spectroscopy. The laser produces extremely "pure" radiation and is capable of providing extremely detailed information about the interaction of electrons in atoms if the color of the laser is suitably chosen. Schawlow developed a means whereby lasers with this very pure output radiation could selectively be tuned to interact with atoms. The precision of this method is so great that it can be used to experimentally test for the first time fundamental theories about matter itself.

Established by Congress in 1946, ONR is the oldest scientific research contract agency in the federal government. The Contract Research Program, which ONR manages, not only provides compensation for university scientists but also provides funds for university research facilities and equipment. Including Professors Bloembergen and Schawlow, ONR has supported 25 Nobel Laureates in the fields of Physics, Chemistry, and Physiology or Medicine during its 35 year history. ■



Electrically Induced Regeneration

by

Stephen D. Smith
University of Kentucky

Introduction

Unfortunately, people at times lose fingers, arms and legs in various types of accidents. Such disasters leave the victims at a loss physically and financially. Medical expenses as well as lost income and decreased personal productivity can be overwhelming obstacles.

The manufacturers of artificial limbs (prostheses) can provide functional replacements of limited utility. However, the artificial limbs of today require expensive prostheses-fitting, as well as extensive retraining of the person in order to take a useful place in society. These problems could be greatly helped if lost limbs could be regenerated by an individual's own body. This is the aim of our current research—to replace lost parts with what, in essence, is original equipment. That involves growing back parts such as arms and legs which are comprised of numerous different component tissues such as bone, nerve, muscle, and blood vessels. Our research method is to use minute amounts of electricity to stimulate the remaining cells in wounded areas to first reproduce themselves by replication and then to duplicate the lost part by the process of differentiation. The initial phase of this research, which is described in this paper, has been accomplished with the support of the Office of Naval Research.

Dr. Smith is a research scientist and Associate Professor of Anatomy at the University of Kentucky College of Medicine. His research interests lie in the field of the effects of physical/chemical agents on growth and differentiation of cells and organisms. He is the recipient of numerous research grants, and has published extensively in the field.

History

Bioelectrical investigations were dramatically brought to public attention in the mid 1700's by Luigi Galvani, who found that when muscles contract, a tiny electrical current is generated. He also discovered that muscle contractions can be stimulated with an electrical shock—galvanize it into action, so to speak. This observation led to a variety of esoteric theories on the role of electrical currents in the body.

A solid scientific approach to the study of biologically significant electrical events during growth and differentiation was rekindled in the first part of this century by A.P. Matthews[1]. In 1903 he discovered that living hydroids (small relatives of sea anemones) are electrically polarized—that is, the "head end of the animal, with the flowerlike hydranth, has a different electrical potential than the "foot" end, which is fastened to rock. This results in a measurable voltage difference along the length of the animal. Subsequently, E. J. Lund[2] reasoned that if such electrical fields are present in animals (and, incidentially, plants as well), they might have something to do with the structural organization and development of the bodies of the organisms in question. He tested this by observing growth of eggs of the seaweed *Fucus* in seawater in the presence of an applied current created by placing electrodes at the opposite sides of a petri dish. The eggs always produced their first outgrowths directed toward the negative electrode. He then demonstrated that animals also respond to electrical fields by showing that he could control which end of a piece of hydroid would become the head and which the foot by applying appropriate fields. This rather surprising results has been confirmed many times by many others, and is now a well-established phenomenon.

Since that time, it has been demonstrated in numerous laboratories that the polarity in growing systems is amenable to manipulation by the application of electrical fields. We have done it with protochordates (primitive cousins of vertebrates), and it has been shown for flatworms as well. The real question of course, is whether this type of control can be exercised in the case of higher vertebrates. The answer seems to be a qualified yes, at least so far.

As early as 1920[3] it was shown that chick nerve cells cultured *in vitro* would survive and send out nerves toward a cathode when a small D.C. current was applied to the culture dish. For technical reasons, the validity of these and similar results were questioned, so we repeated them in 1975[4] under better-controlled conditions, and confirmed the earlier findings. In addition, we found that the nerves not only lived and grew out toward the cathode, but that they grew two or three times faster than normal. This suggested that a small D.C. current, if applied to an injured and repaired tissue might cause better

regeneration than otherwise in two respects: (1) the speed with which the outgrowing fibers crossed the gap into the severed segment, and thus the speed of return to function, and (2) a more accurate reconnection of the severed fibers to their original pathways. Furthermore, the current may serve to direct reinnervation in a more organized fashion.

Research Today

The question is, of course, how to apply a current to nerves in the body. One can easily run wires into a culture dish, but it is not so easy to put them into whole animals. Wires leading through the skin can result in infections, and the movements of the animal or person may displace or break the metal. We used a technique employed earlier in some experiments for regeneration of frog limbs. We decided to let the animal be its own battery and implant the whole device beneath the skin. The method is illustrated in Figure 1, and has been carried out by our colleague, Dr. William G. Winter, at the Denver V.A. facility. In brief, a rat is anesthetized, and its sciatic nerve (the large nerve in the back of the leg) is cut, then sewed back together with very fine stitches. Dr. Winter then inserts the ends of a bimetallic couple into the nerve on either side of the cut. The couple is made by fusing a small (30 gauge) platinum wire to an equally small silver wire, then insulating the joint and the rest of the wires down to their tips. When this silver-platinum couple is immersed in the salty body fluids, it generates about .3 volt. The amperage depends on the surface area of the exposed electrode metal, and is adjusted to about 1 to 5 microamperes. This sort of battery is fundamentally similar to the electrolytic one obtained when dissimilar metals are immersed in seawater, and which requires that a piece of "sacrificial" metal be attached to a ship to prevent electrolytic corrosion of vital parts. When such a device is placed in the nerve, the regrowing fibers are attracted to the cathode (silver), and rapidly cross the cut surface. Our results to date indicate that this technique increases the number of fibers that cross the gap, and that the crossing proceeds at a more rapid rate than in control animals whose nerves have simply been cut and reattached. Thus, we anticipate that more rapid and more complete recovery can be anticipated after nerve injuries using this technique.

It is not without faults, however. Two questions immediately suggest themselves. Is it safe to leave wires sticking in nerves over a long period of time? In addition, how do you keep the nerves growing fast once they have passed the end of the implanted wire? Do you have to keep putting in new wires farther and farther down the nerve to keep the regeneration moving along? A partial answer to the problems seems to have been found recently. The experiments of Dr. Arthur Pilla[5], as well as those in our laboratory, have indicated that tiny electrical currents (about 1 microampere per square centimeter) generated as a result of passing a pulsing magnetic field through tissue can substitute for a D.C. field in stimulating the growth of cells. Thus, we may need the D.C. fields only briefly to get the regrowing fibers started in the right direction. Then we can keep them going with a pulsed magnetic field which can be applied externally, with no need to invade the body further. Those trials are proceeding at the present time, and the initial results seem very promising. It may well be that we can in-

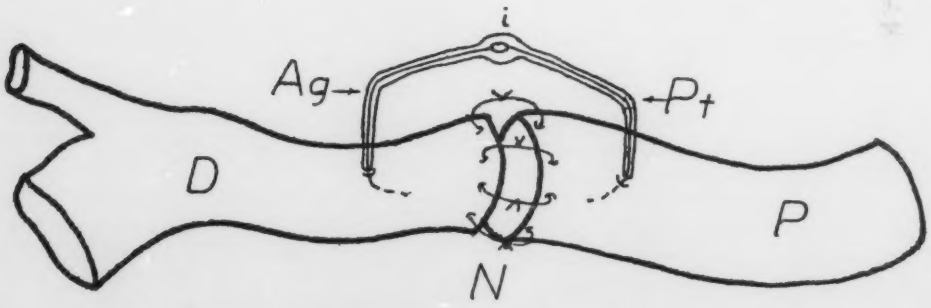


Figure 1. Schematic of nerve repair method. The silver (Ag) end of the implant (i) is inserted into the distal (D) segment of the injured nerve. The platinum (Pt) end is inserted into the proximal (P) segement. The ends are approximately 1 cm. apart across the neurorrhaphy site (N).

sert a D.C. device for a very short time, then remove it and depend on the pulsed fields for the rest of the treatment. In any case, progress is being made on the very difficult task of obtaining good nerve regeneration after a peripheral nerve has been severed. A photograph comparing control and treated (electrically regenerated) nerves (Figure 2) is shown here to demonstrate the improvement in electrically regenerated nerves.

What about the loss of a whole organ, such as an arm or leg? That involves an intricate spatial and temporal array of several different tissues. Not only do we have to arrange which type tissue, how much of each to grow, but also how to make them assume the correct relationships to one another. If we had to do it all by surgical manipulation, it would presently be impossible. We simply do not know enough about limb regeneration, for example, to program the replacement of a lost member. Fortunately, we seem to have the help of the afflicted individual's own body in keeping things sorted out in the right order. And we know that electrical signals are somehow involved, or at least can be involved.

Back in the early 60's, Dr. Robert Becker[6] discovered that the electrical behavior of an amputated frog's limb was different from the electrical behavior of a newt's limb. That was of some interest, since frogs' arms are no better than ours when it comes to regenerating, while newts grow new ones so fast that they scarcely seem to miss them when they are cut off. In fact, a newt has something you can recognize as a regenerating arm within a month after amputation. The immediate thought arises that the electrical difference may be related to the difference in capacity to regenerate. We wondered what would happen if we imposed the electrical pattern seen in an amputated newt's limb on an amputated frog's limb. Accordingly, we implanted a bimetallic couple, of the sort described above for the nerve regeneration experiments, into the limb stump of an amputated adult frog's forearm. For in fact, the frogs grew regenerates which, though certainly far from perfect limbs, contained recognizable organized limb structures. Furthermore, they only did so if the cathode was at the wound surface. In 1967, when the results were published[7], we did not realize how extraordinarily fortunate we had been to have stumbled upon just the right voltage and amperage to produce the desired result. We now know that the threshold for this effect is very narrow, and that we were exceedingly lucky.

We then wondered why the regenerates were less than perfect. Assuming that the reason was related to electrical properties, and perhaps in the position of the electrode at the end of the limbs, we devised the scheme illustrated in Figure 3. We exchanged the bimetallic couple for a battery-resistor circuit delivering a controllable amount of current (about 10 nanoamperes per square millimeter). We also implanted the electrodes in specific sites in one of the four quadrants of the limb stump illustrated in the figure. As it happened, when the negative electrode was placed in the quadrant corresponding to the back of the arm and the little finger side (dorsal/postaxial), remarkably good regeneration ensued. In fact, on at least

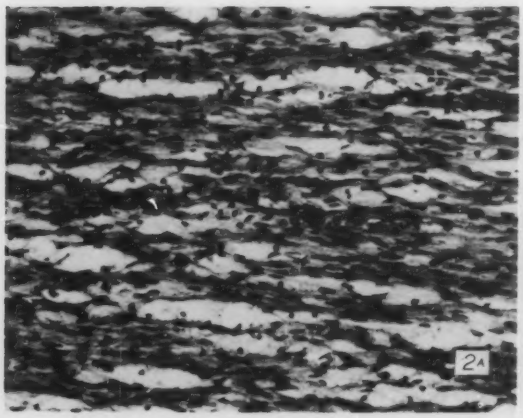


Figure 2.a. Photomicrograph of a regenerated control nerve. Note the many large, degenerated axon sheaths, and the rather chaotic organization of the nerve. Very few normal-looking fibers are present.

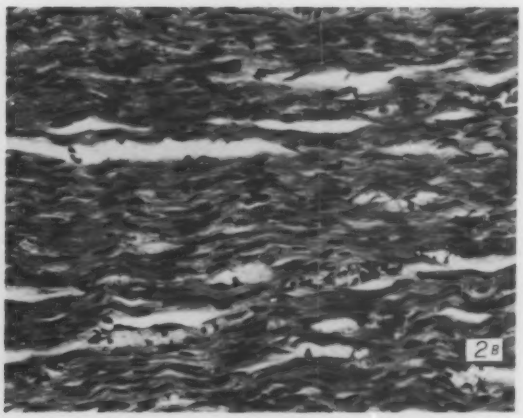


Figure 2.b. Photomicrograph of an experimentally treated nerve. The nerve is much more nearly normal in morphology. There are few axon sheaths showing fatty degeneration, and the fibers are well organized into parallel bundles.

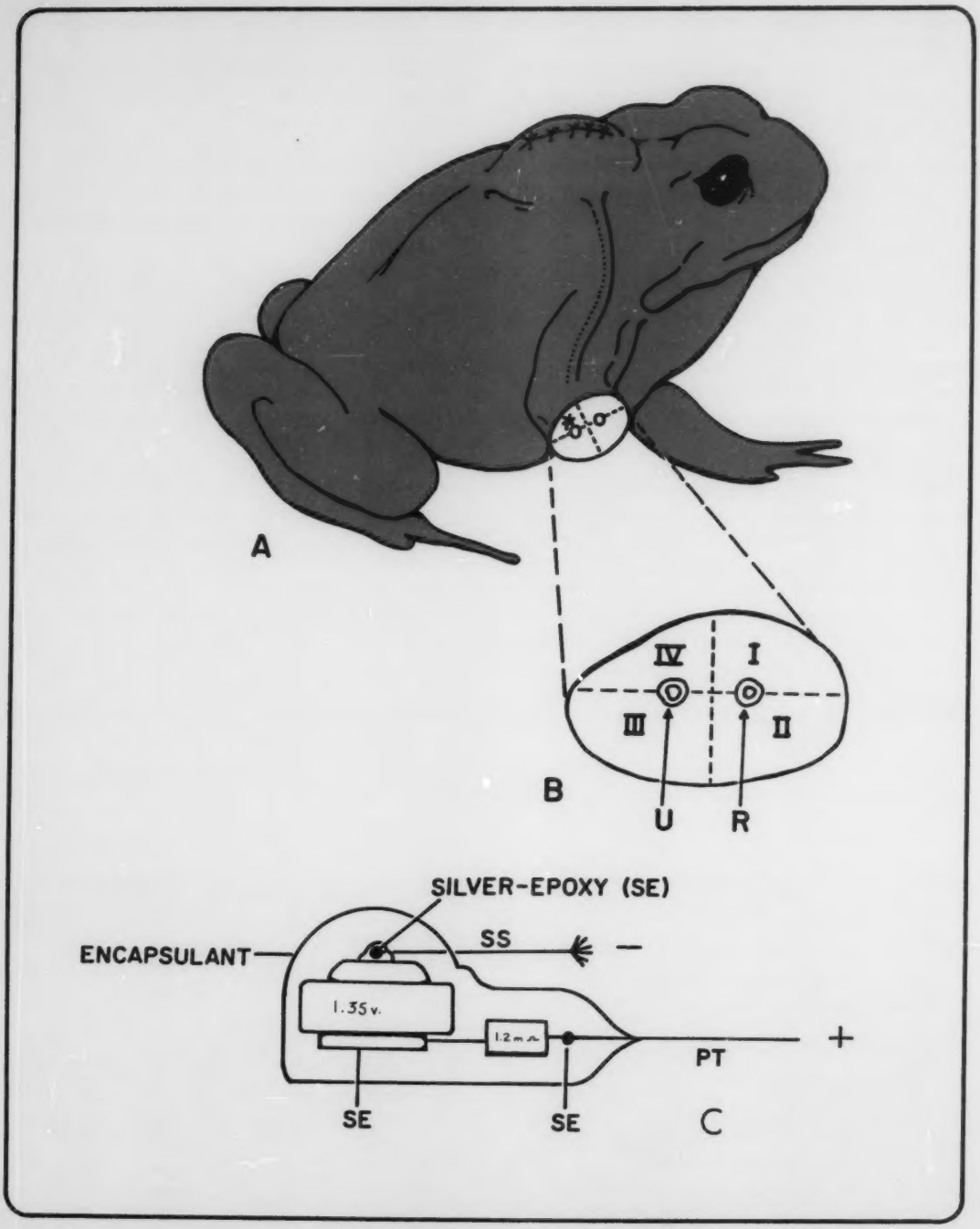


Figure 3. Schema of the operative procedure for regeneration of adult frog limbs. The battery-resistor device shown in (C) is implanted into the back of the frog (A). The negative lead (SS) is threaded down under the skin to quadrant IV at the wound surface (B).

one occasion, a perfect limb grew back.[8] This position of the electrode corresponds to the position of the polarity-determining tissues during the original formation of the limb. In the intervening years, it appears that the answer may have been found by other investigators working on salamander limb regeneration. It seems that for animals who normally regenerate limbs, the limb possesses a "road map" of positional values which lets every cell know where it is in the limb. The coordinates are threefold: one set tells where the cell is around the circumference; a second carries information as to whether the cell is near the surface or near the center of the mass; and a third sets the position of cells along the proximo-distal axis (shoulder-to-fingertips). Thus, every cell has three bits of positional information which precisely define its location in the limb.

With this knowledge in hand, together with our previous results in frogs, we set about to try to repeat our frog results in adult rats. Accordingly, we implanted battery-resistor packs into subadult (60 day old) rat limbs after amputation (Figure 4). As may be seen, these experiments were fundamentally similar to our earlier trials in frogs. So far, we have achieved rather surprising results, but no perfect limbs. Figure 5 shows the best we have been able to do. As can be seen, there are new bones, muscles, joints, and so on, and they are organized into structures that are clearly limblike. However, it is also clear that what has regenerated so far is certainly not a new hand by any stretch of the imagination. It is considerably more than has been achieved elsewhere or before; so it seems we may be on the right track. It may be that in the near future, at least partial regeneration of limbs

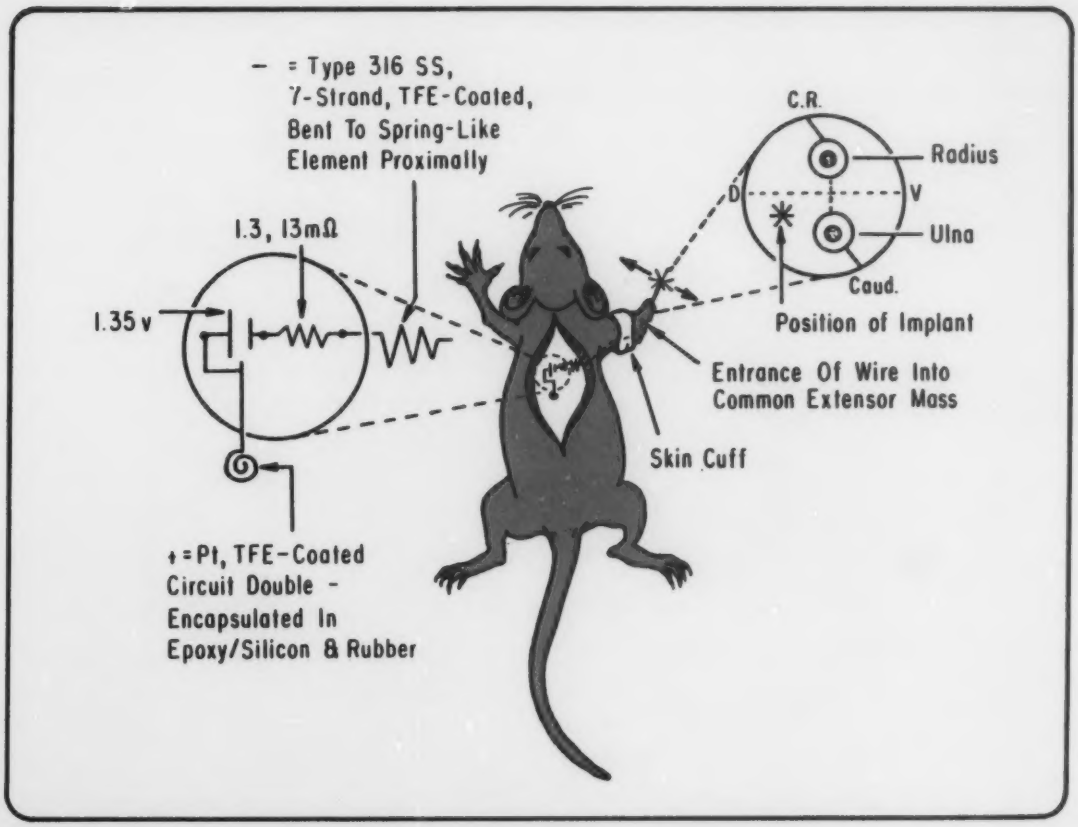


Figure 4. Schema of the operative procedures for regeneration of adult rat limbs. The procedure is obviously essentially identical to that used in frogs.

in adult humans may be possible.

We assume that the following steps will be involved:

1. The body must know that the part is missing;
2. Enough cells must be generated to make a new part;
3. The new cells must have a map so that they know what to become; and
4. They must be set on the road to doing so.

Currently, it appears that condition one is normally present. Conditions three and four are apparently satisfied or reinforced by the application of D.C. fields. We think condition two is the sticking point at present. It may well be that there just aren't enough new cells present to make a complete limb when the time for differentiation arrives. Thus, an incomplete one is tried. It may be that we shall have to be satisfied with a less than complete new limb. We are now combining treatment by D C. fields (to provide the map and starting switch) with stimulation by pulsed magnetic fields in an attempt to increase the number of newly formed cells available for limb formation. Since those experiments are just now in progress, we don't have the answer yet. ■

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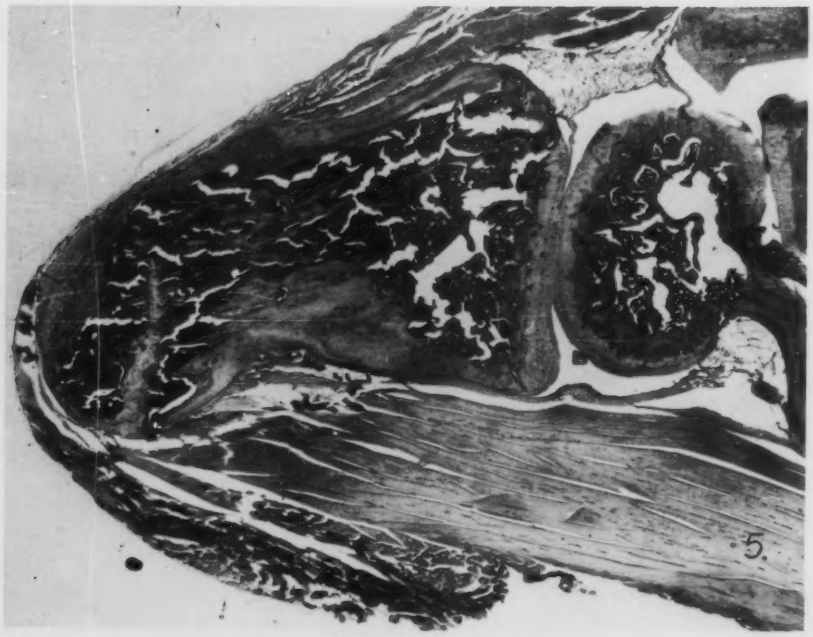

Figure 5. A photomicrograph of a partially regenerated rat limb. There is new bone, muscle, cartilage, tendon, ligaments, etc., and the arrangement of tissues is strongly reminiscent of normal wrist structures.

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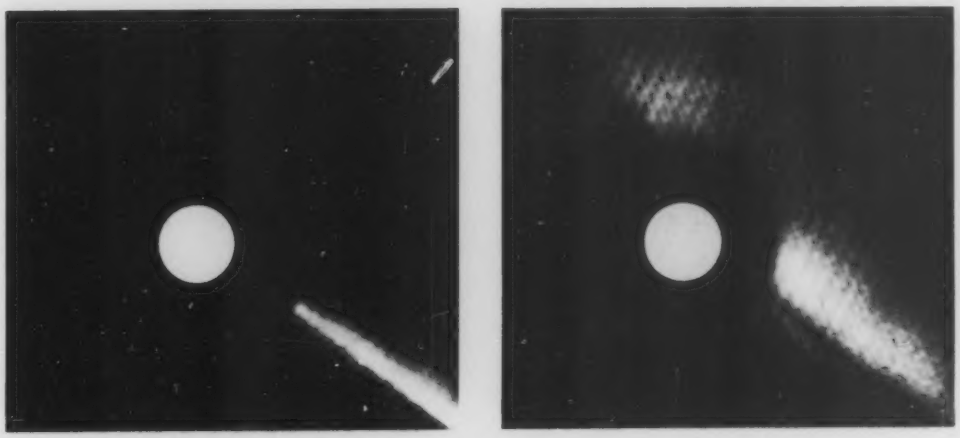
NRL Reports Comet Collides With Sun



In the left photo, obtained by the SOLWIND coronograph, the comet is seen streaking toward the sun at a speed of about 640,000 mph, trailed by a tail of dust and gas more than three million miles in length. On the right, the glow surrounding the disk indicates a dust storm caused by the comet collision.

Scientists at the Naval Research Laboratory (NRL), report that on 30 August, 1979, a comet collided with the sun, scattering cometary debris in an immense halo that rose millions of miles above the solar surface.

This event, according to NRL researchers, marks the first positive evidence of a celestial body colliding with the sun, and is the first instance of a comet being discovered by satellite instrumentation.

"The discovery occurred quite by chance," states Dr. Donald J. Michels, Chief Scientist for NRL's SOLWIND experiment, the instrument that recorded the event. SOLWIND is operating in earth orbit aboard the Department of Defense Space Test Program's P78-1 satellite.

Because of delays in the release of the satellite's data to the NRL experimenters for analysis, the comet/sun collision was not discovered until recently.

The SOLWIND experiment contains a coronagraph designed to monitor activity in the sun's outer corona. It provides a view of the corona similar to that seen from the ground during a total solar eclipse. To do this, the coronagraph has its own "artificial moon," or occulting disk, which provides a continuous "solar eclipse" for the instrument. "Total eclipses observed from the earth last no more than a few minutes," says Michels, "while SOLWIND has been able to observe the sun's corona through these artificial eclipses night and day for nearly three years."

According to Michels, the comet passed through the instrument's field of view as it streaked on its collision course with the sun. It quickly disintegrated upon impact in the sun's blazing heat.

Electron Device Reliability

by

Max Yoder
Office of Naval Research

The performance and reliability of electronic systems has been and continues to be of paramount concern to the Navy. For the past two decades there has been a concerted effort to replace vacuum tubes with solid state devices which are much smaller and not thought to possess the built-in wear-out mechanisms of tubes. In an attempt to increase performance, much engineering effort has been devoted to squeezing the last available erg of energy from solid state devices, particularly in the field of solid state microwave amplifiers and sources. This "squeeze" has created devices that operate with higher electric fields and at higher temperatures. The high performance devices thus developed, however, are frequently less reliable than the vacuum tubes they are intended to replace. Although many reliability improvements have been made in recent years by using refractory metals with gold overlays for all on-chip interconnections, a built-in wear-out mechanism still exists. Although its symptoms are well-known, its underlying fundamental cause has evaded conclusive understanding.

Dr. Thomas Magee of the Advanced Research and Application Corporation (ARACOR), under Office of Naval Research support, recently made a dramatic discovery regarding the failure mode of gallium arsenide (Ga As) power field-effect transistors (FETs). Using a transmission electron microscope (TEM) and spatial distributions of chemical concentrations obtained from a secondary ion mass spectroscopy (SIMS) technique, he examined numerous Ga As microwave power FETs that had failed during life tests while operating at full bias and with radio frequency (RF) gate drive. What he found was that copious quantities of chromium (Cr) impurities had migrated to the vicinity of the positive drain terminal as shown in Figure 1. This was the first conclusive proof that impurity diffusion in an operating Ga As microwave solid state device is not gaussian (random), and that migration of impurities does occur toward regions of highest electric field strength such as near the positive drain contact. As the Cr ions migrate towards the positive drain contact their concentration there increases until, at a concentration near 10^{20} parts per cubic centimeter, the device fails. As the Cr concentration increases, the concentration of holes (electron traps) increases proportionately. Electrons in the FET channel must traverse this hole-abundant region on their way to being collected by the drain contact. In attempting to pass this region, they can be trapped by the holes with emission of light in a process known as recombination radiation. Thus it is quite plausible that the recombination radiation typically observed near the drain contact of a power FET at incipient failure is induced by the chromium migration into the region of the drain contact.

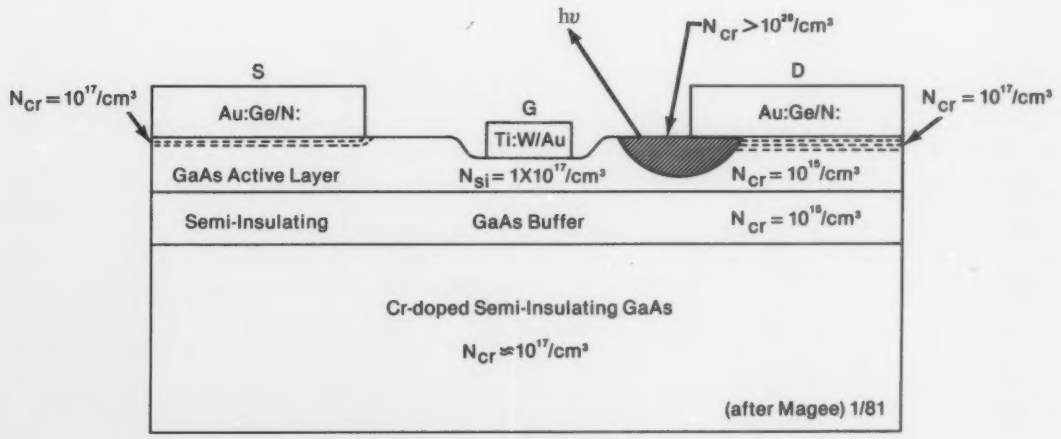
This discovery no doubt describes the principle cause of wear-out mode failure in high power Ga As

microwave FET devices. Clearly, a method is needed to prevent the chromium migration towards the high field region. One obvious approach is to eliminate the chromium impurity altogether. Unfortunately state-of-the-art technology does not allow this approach. While the chromium can be reduced to concentrations approaching $10^{13}/cm^3$, this approach merely delays the inevitable, for even at low background concentrations the chromium will eventually accumulate at the drain contact.

To design greater reliability into Ga As power FET devices will require the incorporation of a mechanism that "ties-up" the background chromium to physically prevent its migration to the drain contact. Such an approach appears promising. In a recent paper presented at the Semi-Insulating III—V Materials Conference,[1] it was proposed that impurities in Ga As can be made to complex with each other and that the resulting complex would be too large to migrate or diffuse through the crystal lattice. Thus, we need an impurity that will complex with chromium in the Ga As lattice and will not otherwise interfere with device operation. Candidate impurities that also are suitable for creating the required donor impurity levels in the active channel regions include tin, sulfur, and selenium. Indeed, tin has recently been found to complex with chromium in a Ga As host crystal[2].

The successful application of the complexing approach to the reliability problem may also engender a serendipitous enhancement of Ga As power FET performance. This may be better understood by examination of Figure 2 which shows the impurity profile of a tin-doped active FET channel together with profiles of the background chromium and of the chromium after a long period of field assisted diffusion. The background chromium within the active tin-doped channel is complexed with the tin and does not migrate whereas the background chromium beneath the tip-doped channel region begins to diffuse toward the semiconductor surface in the direction of the drain contact. As it approaches the region of tin doping (backside of the conduction channel of

Mr. Yoder is an electronics engineer in the Electronics Division of the Office of Naval Research since 1962. He is well known for his many patents and publications in the research areas of electronic materials, microwave devices, and gigabit logic.



Spatial Distribution of Chromium Concentration after GaAs Power FET Failure

Figure 1.

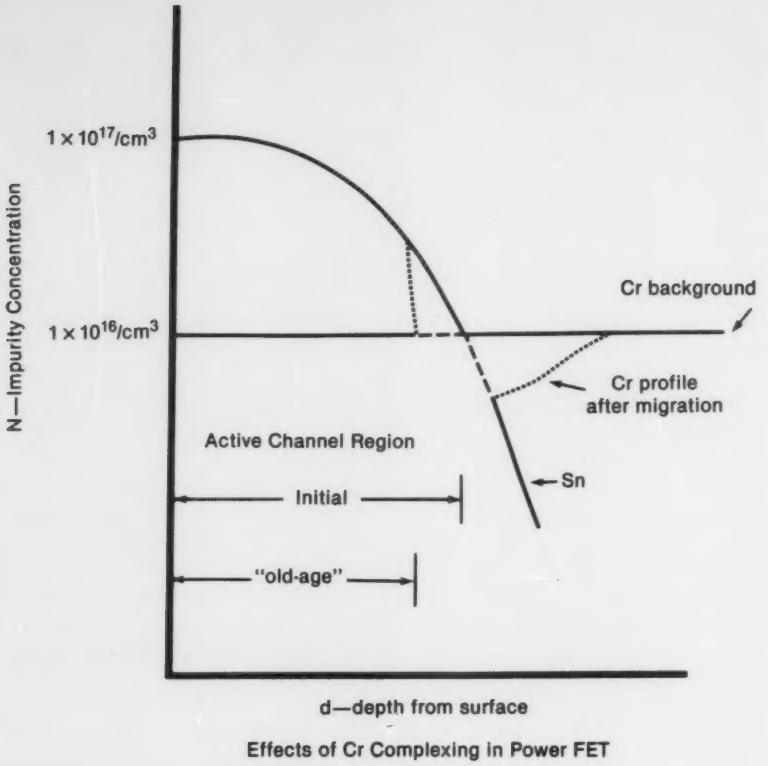


Figure 2.

the FET) it complexes with the tin and builds up to concentrations much greater than the background Cr level, but less than the maximum tin concentration level. The limitation of maximum Cr concentration levels (much less than $10^{20}/cm^3$ immediately adjacent to the drain experienced in conventional designs) results from the complexing action occurring at distances removed from both the immediate region of maximum electric field strength and the immediate region of maximum crystal strain induced by the formation of the drain contact. The device performance enhancement results from the fact that some Cr diffusion into the tin-doped active channel region does occur with the result that the electrical donor activity of the tin is compensated or cancelled-out. This limited compensation results in a much sharper fall-off (with depth) of the electrical conductivity profile of the active channel. As a result, the device pinch-off characteristics become much sharper and the device output conductance improves. Figure 3 illustrates the electrical conductivity profile both before and after Cr migration. It can be seen that the device performance actually improves with operation as compared with the more familiar condition of degradation with age.

The complexing of the chromium in Ga As represents but the first step in understanding and using complexes in compound semiconductor materials. Recent findings indicate the possibility that the *majority* of shallow level electronic activity in compound semiconductors may accrue from impurity complexes rather than from the simple hydrogenic model inherited from elemental semiconductor theory. A broad new area of semiconductor science may just be beginning to unfold. ■

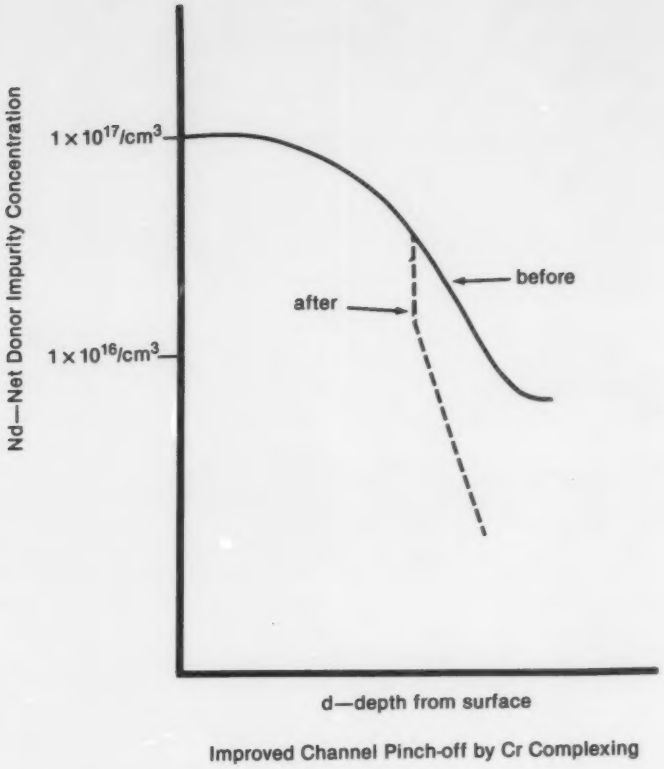


Figure 3.

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research notes

Another Significant Submicrometer Lithography Advance

Semiconductor device performance advances in recent years have been inversely proportional to dimensional tolerances achievable in fabricating these devices. Breaking the 1 micrometer barrier required the introduction of expensive x-ray and/or electron beam lithography equipment. At 0.2 micrometer resolution an unexpected problem was encountered. These extremely small openings in the "resist" material simply did not provide enough "ventilation" to outgas trapped vapors. While another expensive process—ultra high vacuum—solved this unexpected problem, another unexpected problem arose.

Semiconductors require metallization paths to carry signals from one active device to another and to the "outside world." Metallization of openings less than 0.2 micrometers wide by conventional metallization techniques, however, encountered another new problem. The opening in the top of the resist would close over before the underlying hole was full. This unfortunate result was faithfully reproduced by many frustrated investigators regardless of the metal used, the deposition rate used, or the type of deposition (e.g., evaporation or sputtering). The result of this pinch-off or close over of the resist opening was that the metallization could not be made high enough to provide enough cross sectional area for the metal so as to provide adequate electrical conductance to the underlying device. Electroplating, while building up the thickness of the deposited metal, unfortunately builds up the width also with the loss of lithographic resolution so tenuously achieved. A double resist masking step subsequent to first deposition and prior to electroplating which, in principle, may work, requires a realignment resolution not currently reproducible.

The basic problem resolved into one or more of three possible causes. They are (a) the "rolling" of atoms as they impinged the substrate, (b) lateral energy of the evaporating (sputtering) metal and (c) deposition rate. The "rolling" problem was approached by distinguishing among (1) atoms with spherically symetrical S-type outer electron shells and (2) highly asymetrical d-type outer electron shell configurations. In practice, there appeared to be no difference between metal types or deposition rates. The first attempt to "collimate" the evaporated metal beam to reduce lateral molecular energy was only marginally effective. A 4-stage collimator, however, met with remarkable success. It provided a molecular metal beam of such laminarity that metals can now be deposited into slits as small as 0.15 micrometers and filled all the way to the top of the resist layer without experiencing close-off. This result should enable the reproducible fabrication of low noise field effect transistors operating well into the milimeter wave spectrum and characterized by large associated gain.

The above work was accomplished at Varian, under ONR contract. Varian, for three consecutive years, has demonstrated world records in low noise, high associated gain microwave amplifiers. This important advance in submicrometer device metalization technology should further improve these results. (Max N. Yoder, ONR) ■

Photoemission Spectroscopy

The monochromatic storage ring excited photoemisson of electrons from surfaces was shown by G. Magaritondo of the University of Wisconsin to be sensitive to an electron escape depth of about 4 to 6 Angstroms for 50 to 100 eV electrons. This new spectroscopy has been demonstrated to be useful for several new types of measurements: (1) A comparison of core level excitation energy in silicon and germanium revealed a discrepancy of 0.3 eV and 0.35 eV respectively when measured by photoemission

and by electron loss spectroscopy. Because electron loss spectroscopy is sensitive to a fraction of a layer of adatoms, this discrepancy is interpreted to be a shift in the core level between an atom in the bulk and an atom on the surface. (2) Combined with low energy electron diffraction Al evaporated onto GaAs was shown to displace the Ga atoms not in the first but in the second layer of the (110) surface. Subsequent annealing results in the Al atoms diffusing deeper into the bulk of the GaAs. (3) The thickness of the interface formed by metal deposited onto a III-V semiconductor varies inversely with the strength of the bond formed between the metal and the semiconductor. For example, Al on GaAs has a 10 Angstrom interaction width while Cu on GaAs has an interaction width of 100 Angstroms.

The usefulness of photoelectron spectroscopy at revealing the atomic geometry of interfaces and vertical position of metallic atoms deposited on semiconductors is of enormous importance in the fabrication and life of microelectronic circuits. L. J. Brillson of Xerox and A. Kahn of Princeton are collaborators in this work, which is supported by the Office of Naval Research.
(Van Nicolai, ONR) ■

A New Insight Into the Origin of Dolomite on the Ocean Floor

Carbonate rocks are very common in the geological column. Among the minerals composing these rocks three are prominent: calcite ($CaCO_3$), aragonite ($CaCO_3$), and dolomite ($CaMg(CO_3)_2$). It came therefore as a major surprise when it was found many years ago that in sediments formed in the present ocean calcite and aragonite are widespread, but dolomite is exceedingly rare, although thermodynamic calculations have shown dolomite to be the most stable carbonate mineral in sea water.

Many attempts were made, particularly during the last thirty years both for scientific and economic reasons, to explain the scarcity of recent dolomites; rocks composed of dolomite are generally much more porous than those built of calcite and/or aragonite, and are therefore in many areas of the world reservoir rocks for oil. Most sedimentologists assumed so far that the formation of dolomite is mainly controlled by the dissolved magnesium/calcium (Mg^{+2}/Ca^{+2})-ratio in sea water. As the molar Mg^{+2}/Ca^{+2}-ratio in sea water is 5.3, it was assumed that dolomite formation requires a still higher ratio. These explanations, however, were contradicted by the observation that in a few places in which modern dolomite is being formed, e.g., the coastal flats of the Persian Gulf, the Mg^{+2}/Ca^{+2}-ratio in the interstitial water is not particularly high.

Experiments conducted recently in the laboratory of Dr. Mariam Kastner of the Scripps Institution of Oceanography have shown that this earlier research into the genesis of dolomite missed the crucial point. The formation of dolomite is not controlled primarily by the Mg^{+2}/CA^{+2}-ratio, but by another of the major components of sea water, namely dissolved sulfate, which inhibits the formation of dolomite. In its absence, sea water reacts rapidly with both calcite and aragonite to form dolomite. In their experiments, both calcite and aragonite in contact with sulfate-free sea water were found to transform into dolomite within a few days to a few weeks, depending on the experimental conditions. The transformation of aragonite to dolomite occurred when the sulfate content of the artificial sea water was less than 15% of its concentration in normal sea water, whereas the conversion of calcite, the most common carbonate mineral in deep-sea environments, to dolomite is inhibited already by traces of sulfate. It was thus concluded that dolomite could form today, wherever sulfate is removed from sea water. The most common process for such a removal of sulfate from sea water is its bacterial reduction to H_2S within marine sediments rich in organic matter. Environments like these occur in many near-shore basins, such as the Gulf of California or the California Borderland, and in these sediments the present formation of dolomite was in fact lately discovered.

The discovery of the inhibiting effect of sulfate on the formation of dolomite appears to be the clue for an understanding of dolomite genesis in nature. One conclusion might well be that dolomite formation in recent marine sediments close to the sediment/sea water interface is more common in near-shore submarine basins than was hitherto assumed.
(Mark B. Odegard, ONR) ■

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